BITTER CRAB DISEASE STUDIES: OBSERVATIONS ON SEASONALITY, MORTALITY, SPECIES SUSCEPTABILITY AND LIFE HISTORY

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ABSTRACT

Incidence and average intensity of Bitter Crab Disease (BCD) in Auke Bay Tanner crabs (Chionoecetes bairdi) were significantly greater during June through September of both 1989 and 1990 then during October through May. BCD is a chronic but fatal disease; crabs did not develop immunity and often died from secondary bacterial and ciliate infections. Total mortality exceeded incidence and was not significantly different between summer and winter seasons or between years. BCD appears to be host specific: red king crabs (Paralithodes camtschaticus) and Dungeness crabs (Cancer magister) did not contract BCD post-injection. BCD amoeboid stages consistently caused disease in Tanner crabs when injected into the hemocoel, while dinospore stages did not. Waterborne challenges did not cause disease. BCD parasites did not occur intracellularly, remaining within the hemal and vascular systems. Parasites exited the host via gills and possibly esophagus. The life cycle of BCD dinoflagellates outside their hosts remains incompletely described.

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CHAPTER I.

BITTER CRAB DISEASE STUDIES: AN INTRODUCTION TO THE LITERATURE

The Tanner crab (Chionoecetes bairdi Rathbun, 1924) and snow crab (Chionoecetes opilio Fabricius, 1788) fishery in Alaska is a multi-million dollar industry and the primary invertebrate fishery in Alaska. In southeastern Alaska during the 1988-1989 commercial season this industry netted about $3 million (Imamura and Botelho, 1989). The Bering Sea component of this fishery is vastly larger.

Since 1985, an increasing number of Tanner crabs from southeastern Alaska have been found to be infected with a parasitic dinoflagellate similar to Hematodinium species. This species of parasite is known to infect the blue crab (Callinectes sapidus), and several other species of western and eastern Atlantic crabs. In 1988 and 1989 a number of Tanner crabs (C. bairdi) and snow crabs (C. opilio) from the Bering Sea were found to harbor the parasite (Meyers et al., 1990). The syndrome has been named Bitter Crab Disease (BCD).

Infected Tanner crabs are pink in color, have milky hemolymph, chalky-textured meat with a bitter aspirin-like aftertaste, and are unmarketable. Fishermen and processors alike have been aware of occasional chalkiness and bitter flavor in Tanner crabs for several years. Most of these

tainted crabs are sorted on the fishing grounds or at the processor's docks. The practice of discarding infected crabs overboard may have facilitated the spread of the disease throughout its present range, both in southeastern Alaska and possibly the Bering Sea. Some areas in southeastern Alaska have an incidence of infection approaching 100 % (Eaton et al., 1991) and have been voluntarily closed to commercial fishing. Disease-free areas currently open to fishing could also be closed if BCD were to spread.

Fishermen and processors have suffered significant economic losses in the past few years as a result of catching and purchasing diseased crabs that were subsequently unmarketable. In southeastern Alaska, the Alaska Department of Fish and Game (ADF&G) Commercial Fisheries Division has estimated a loss of over 80,000 lbs of crab worth over $220,000 in 1989 due to Bitter Crab Disease (Cathy Botelho, ADF&G Commercial Fisheries Division, personal communication). In certain areas 40-80% of the Tanner crabs brought to the processors during the 1988-1989 season were unmarketable. Some fishermen report throwing away over 50% of their catches prior to delivery (Kevin O'Sullivan, Alaska Seafood Marketing Institute, personal communication). If these catches were added to the 80,000 pounds of bitter crab inadvertantly delivered to processors, the estimated $220,000 loss would be much higher.

Hematodinium perezi was first described by Chatton and Poisson (1931) in the portunid crabs Carcinus maenas and Portunus depurator off the Atlantic coast of France. In one of the infected crabs, they noted "opalescent" hemolymph and tissues, characteristics probably due to a heavy parasite load. The complete life cycle was not fully described and no flagellar stages were found. Their identification was based on morphological and developmental similarities between Hematodinium and Syndinium, a genus found in copepods which has a flagellated stage.

Additional hosts were not described until Newman and Johnson (1975) found H. perezi in the tissues and hemolymph of the blue crab, C. sapidus in the coastal waters from North Carolina south to Florida. The disease occurred in prevalences up to 30% but was not found from late winter to early spring or in salinities below $11^{\circ}/_{oo}$. As with the European crabs, the tissues and hemolymph were milky-white and moribund crabs were extremely lethargic. Newman and Johnson (1975) did not describe any flagellated forms although unicellular and highly motile, vermiform, multinucleate bodies were seen. In blue crab populations, H. perezi infections of up to 30% may cause significant mortality.

MacLean and Ruddell (1978) extended the parasites' host range to include rock crabs (Cancer irroratus), Jonah crabs (Cancer borealis), and lady crabs (Ovalipes ocellatus).

Infected crabs collected from the inshore and offshore waters of the mid-Atlantic Bight made up 0.6 to 4.0% of any given sample. Infected C. borealis sampled from offshore sites were collected during all parts of the year and no crabs over 7.0 cm carapace width were taken. Two C. irroratus had an unusual color similar to the pinkish-red described by Jepps (1936) in infected calanoid copepods.

Benthic amphipods from the northeastern U.S. are also parasitized by H. perezi. Johnson (1986) described the parasite in thirteen species of benthic amphipods taken from the continental shelf and observed prevalences of infection from <1 to 67%. Unlike Syndinium gammeri described by Manier et al. (1971) which produces a single, massive plasmodia that eventually castrates its amphipod host, the parasite observed by Johnson (1986) produces uninucleate, plasmodial and spore stages, similar to BCD dinoflagellates in Tanner crabs from Auke Bay. Histological examination did not indicate that the amphipod immune system recognized S. gammeri as being foreign. Since hemocytes did not aggregate around or phagocytize dinoflagellate plasmodia but did appear to respond to systemic fungal infections. Amphipod mortalities probably resulted from secondary infections. Heavy parasitism in amphipods may also regulate population size, as is possible in blue and Tanner crabs.

Bitter crab disease (BCD) was first described by Meyers

et al. (1987) in Tanner crabs from southeastern Alaska. In vivo transmission studies by Eaton et al. (1991) indicated possibly nine different morphological forms: five vegetative, two prespore and two spore types. The five vegetative stages require several months to mature and appear to divide into tremendous numbers inside the crab eventually developing into the two prespore types. During this process, most of the normal hemocytes are destroyed and the hemal sinuses fill with parasites. The bitter flavor may come from extracellular products released by the parasite, which are visible under light microscopy as a very foamy cytoplasm and by electron microscopy as droplets exuding from the surface of the vegetative cells (Meyers et al., 1987). The two spore types consist of a larger, slow-moving and a smaller, fast-moving bi-flagellated dinospore. The vegetative stages appear to be only passively infectious and may be excreted or passed with the feces in heavily infected animals. The motile dinospores appear to be diploid and are considered to be infectious (Eaton et al., 1991). Preliminary results on the incidence and intensity of infection near Sullivan Island in southeastern Alaska showed that the lowest level of infection occurred in the fall and winter months and increased in the late winter and early spring. Newly molted crabs were more likely to be infected than old-molt animals (Eaton et al., 1991).

At present, many questions concerning bitter crab disease remain unanswered. The following chapters present the results of three different research questions concerning aspects of the life history of the BCD parastie and impacts of disease. Chapter I addresses the question of the seasonal nature of BCD infection and mortality rates. Chapter II deals with the susceptablity of other commercially important decapods, the red king crab and Dungeness crab, to the BCD parasite. Research summerized in Chapter III addresses questions regarding the BCD parasite's life history as well as the histological effects of severe parasitism. Research of this type provides additional insight regarding the biology and pathogenicity of the the dinoflagellate causing Bitter Crab Disease and aid scientists in appropriate disease as well as host management decisions.

CHAPTER II.

SEASONAL MORTALITY, INCIDENCE AND INTENSITY OF BITTER CRAB DISEASE (BCD) IN ALASKAN TANNER CRABS (Chionoecetes bairdi) FROM AUKE BAY, ALASKA

Introduction

Since the 1984-1985 Tanner crab (Chionoecetes bairdi) season in southeastern Alaska, an increasing number of crabs infected with a dinoflagellate parasite have been identified in the commercial catches. The syndrome caused by the dinoflagellate is known as Bitter Crab Disease (BCD). Total reported deadloss in excess of 80,000 pounds worth about $220,000 during the 1988-1989 season was attributable to BCD (Cathy Botelho, Alaska Department of Fish and Game(ADF&G), personal communication). More recently, Tanner crab and snow crab (Chionoecetes opilio) from the Bering Sea were also found to be infected with this dinoflagellate, although incidence of infection was lower than in southeastern Alaska (Meyers et al., 1987; Meyers et al., 1990). The potential biological and economic impact of this fatal disease could threaten these highly valued fisheries.

Increasing demand for Tanner crab and depressed supplies of king crab have made the harvest of Tanner crab a multi-million dollar industry. During 1989, approximately 164.6 million pounds of Tanner and snow crab were harvested from

U.S. waters. Valued at about $160.1 million, this fishery represented 10% of all crustaceans harvested from U.S. waters by value and 4% of the total fish and shellfish landings. Among invertebrate fisheries, Tanner and snow crabs were second in value only to Gulf of Mexico shrimp during 1989 (O'Brannon, 1990). The Tanner crab (C. bairdi) fishery in southeastern Alaska grossed $4,247,763 during the 1989-1990 commercial season, higher than any other local invertebrate fishery (Cathy Bothelo ADF&G, personal communication). Tanner crab landings have been relatively stable at an average of 1.7 million lbs since the early 1970's providing an important source of income to southeastern Alaska fishermen.

The BCD parasite causes a systemic disease in Tanner crabs, characterized by an astringent, aspirin-like aftertaste during the multiple month residence period within the host's hemal sinus and tissues (Meyers et al., 1987). Heavily infected crabs also have a pink abdomen and carapace, white lines along the underside of each merus and milky hemolymph filled with parasites. Infected crabs were not marketable. During July and August, the resident vegetative forms multiplied and developed into one or both of two different sizes of motile dinospores which were observed exiting via the gills thus killing the crab (Eaton et al., 1991, Love et al. 1992, in press). Preliminary results on the incidence and intensity of infection near Sullivan Island in southeastern

Alaska suggested low levels of infection in wild crabs during the fall and winter months, although incidence below 37% was not observed and consistent monthly samples were not collected (Eaton et al., 1991). Newly-molted crabs have a significantly higher chance of being infected and subsequently killed by the BCD parasite (Eaton et al., 1991) Assuming higher incidence and mortality of newly-molted Tanner crabs, the number of new molt crabs in the population should decrease following a year of high disease prevalence.

Knowledge of the seasonal incidence and intensity of BCD in southeastern Alaskan Tanner crab populations would enable managers to choose seasons for fishing openings so as to minimize harvest of unmarketable, heavily-infected crabs, allow harvest of lightly infected, possibly marketable crabs, and protect local disease-depleted stocks from possible overharvesting. This project was designed to determine the seasonal variations in incidence and intensity of BCD in a relatively undisturbed population of Tanner crab which does not support commercial harvest. This study will also address the relationship between crab mortality and average intensity of infection as well as the relationship between BCD infection and molt stage.

Materials and Methods

Tanner crabs were collected monthly from random locations north and west of Spuhn island in Auke Bay and Fritz Cove from

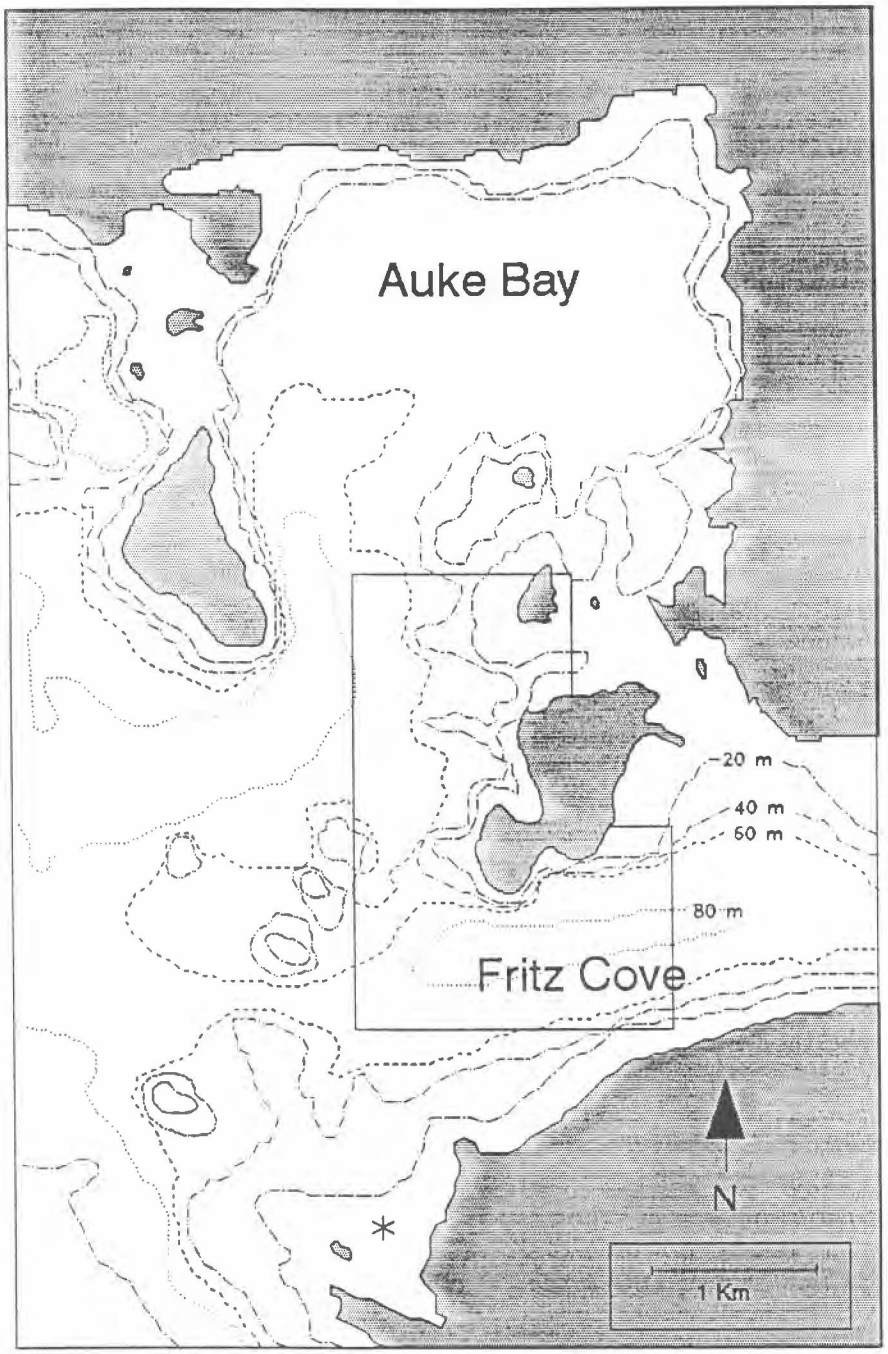


Figure 1. Area in Auke Bay and Fritz Cove, southeastern Alaska, where Tanner crabs were randomly sampled from June, 1989 to September, 1990. Area (asterisk) where newly molted juvenile and female crabs were gathered during April, 1990.

June, 1989 through September, 1990 (Figure 1). Crabs could not be collected with pots in January or April due to mechanical problems with the research vessel but were sampled using SCUBA in April from 10 m when crabs had moved to shallower depths to molt. Crabs were captured by submerging 2m X 2m conical commercial Tanner crab pots for 12-36 hours from waters 90 m in depth. The minimum sample size needed to detect at least one infected crab in populations of 1,000,000 animals or more at a minimal disease incidence of 5%, at the 95% confidence level was determined to be 60 crabs (Ossiander and Wedermeyer 1970). Larger sample sizes would detect infected crabs at lower disease incidence. All crabs were individually tagged with Floy 2 mm fingerling disk tags affixed to the dorsal surface of the carapace with Quick-Gel superglue.

Hemolymph samples from captured crabs were used immediately after collection to make smears on glass slides, which were dried and stained using Diff-Quik histological stain (Baxter Scientific, Inc.). Slides were examined by light microscopy for the incidence and intensity of Hematodinium parasitism. Monthly fluctuations in either could then be followed. All mortalities were assessed for cause of death by dissection of moribund crabs and any releases of dinospores were noted.

Incidence was recorded as presence or absence while

intensity of infection was recorded as per Meyers et al. (1987), with each receiving a numeric code from 0 to 4. Percent incidence and average monthly intensity of infection was calculated monthly.

Crabs were then placed in aquaria and fed a mixture of frozen squid and mussels for an observation period of 60 days. All crabs observed releasing dinospores were examined postmortem. At the end of this observation period a final hemolymph sample was collected and levels of infection were again determined. Newly-molted crabs and old-shell crabs were counted throughout the study.

Incidence and average intensity of infection, sporulation and mortality rates were compared between seasons (summer-fall and winter-spring) and between years (1989 and 1990) using Mann-Whitney non-parametric tests (Zar, 1974). Summer-fall season was defined as June through September and winter-spring as October through May. Incidence of infection and molt stage were compared within months using chi-square analysis. Pooled incidence of infection and molt stage data for both 1989 and 1990 were analysed using two-factor contingency table analysis as described in Zar (1974). χ^2_c was calculated using the following contingency-table formula:

$$\chi^2_c = \frac{n(|f_{11}f_{22} - f_{12}f_{21}| - n/2)^2}{(C_1)(C_2)(R_1)(R_2)}$$

where n=sample size, f_{11}=new-molt +, f_{22}=old-molt -, f_{12}=new-molt -, f_{21}=old-molt +, $C_1=f_{11}+f_{21}$, $C_2=f_{12}+f_{22}$, $R_1=f_{11}+f_{12}$, $R_2=f_{21}+f_{22}$, and $df=(r-1)(c-1)=1$; $\chi^2_{0.05,1}=3.841$, $\chi^2_{0.01,1}=6.635$ (Zar, 1974). Percent presence and absence of disease is denoted by + and -, respectively.

Results

Incidence and average intensity of BCD in Auke Bay Tanner crabs increased during the summer months and decreased to zero by mid-winter (Figure 2). Percent incidence of BCD increased from 75% in June, to 96% in August, 1989, decreased to zero by December and increased again until August of the following year. Incidence of disease was significantly different during June through September (i.e. summer and fall months) then during October through May (i.e. winter and spring) of both 1989 and 1990 ($U=46$, $.001<P<.005$). Percent incidence of infection during August of 1990 was 56%, approximately half that observed in August of 1989 at 96%. Incidence of infection was not significantly different in 1989 from 1990 ($U=25.5$, $P>0.2$). Average intensity of BCD infection in Tanner crab from Auke Bay paralleled incidence, rising until August, decreased to zero by December and increasing steadily again from February until August of the following year. Average intensity of infections was significantly greater from June through September for both 1989 and 1990 than from October through May ($U=49$, $P=.001$). Average intensity of infections

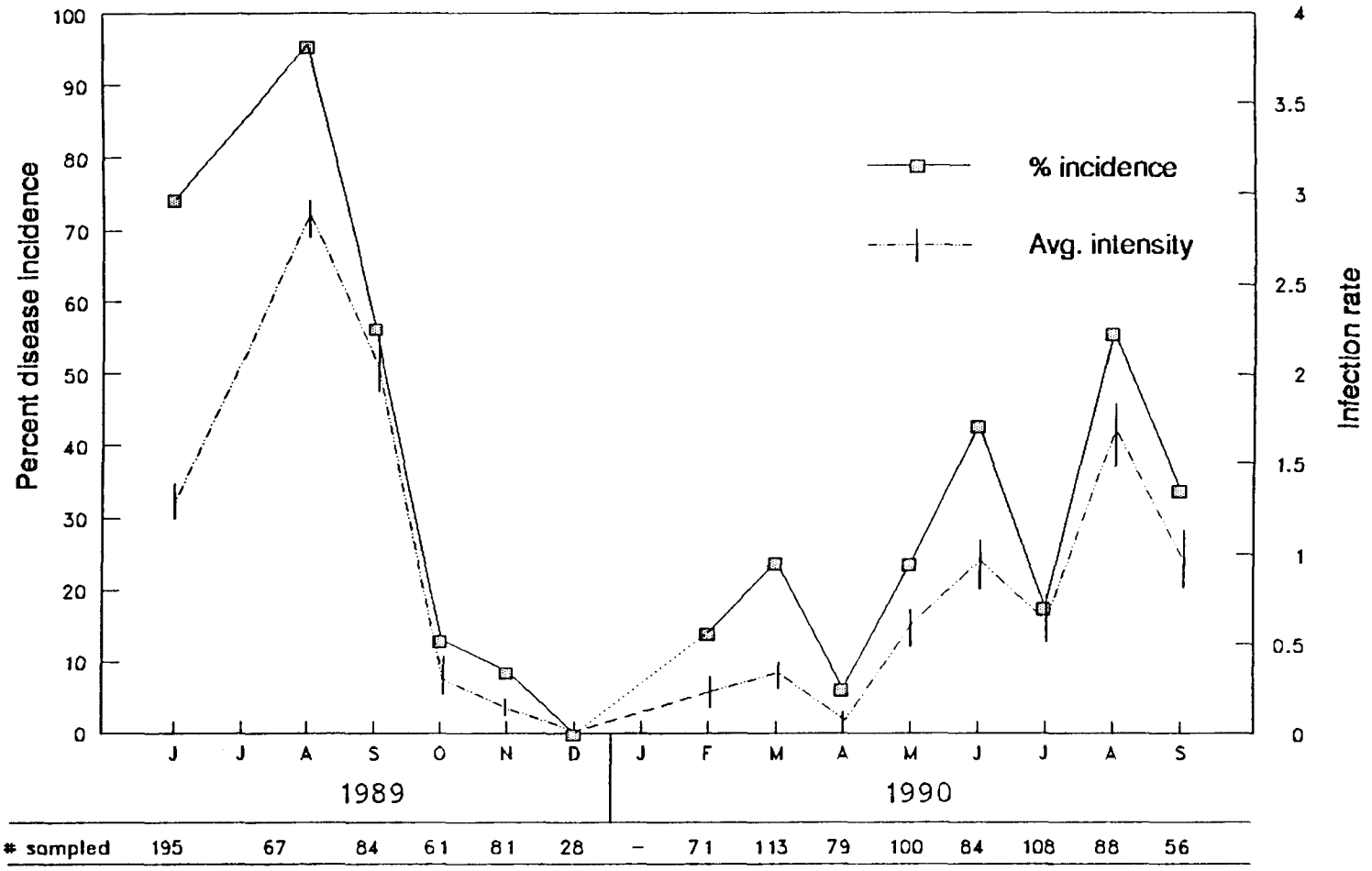


Figure 2. Incidence and intensity of infection with the Bitter crab disease (BCD) dinoflagellate in Tanner crabs (Chionoecetes bairdi) collected in Auke Bay, southeastern Alaska from June, 1989 until September, 1990.

were not significantly different between years (U=26, P>0.2). Sample sizes for all months except December, 1989 and September, 1990 were greater than or equal to 60 animals, allowing disease incidence of 5 % or greater to be detected. Sample sizes in December, 1989 and September, 1990 were large enough to detect disease incidence at the 10% level.

BCD dinospores escaped infected Tanner crabs held in seawater aquaria during the summer months of 1989 and 1990 (Table 1). In 1989, small and large dinospores sporulated from crabs during July through September. During the spring, 1990 dinospores were observed as early as May and continued to exit the crabs until the study was terminated in September. Peak sporulation periods for the large and small dinospores occurred during both years in August and September. Tanner crabs collected during June, 1989 were released immediately following the initial hemolymph sample, and could not be observed releasing dinospores. Most crabs during both years released either small or large spore types, however, on occasion, both dinospore types were produced within the same crab. Total percent sporulation during 1989 was 61.1%, higher than 1990 at 24.9%. Release of BCD dinospores was significantly greater during the summer and fall months than during the winter to spring (U=36, P<.005) but did not vary significantly between years (U'=20.5, P>0.2). Heavily infected crabs which died of BCD but had not been observed

Table 1. Percent of Tanner crabs captured monthly from Auke Bay, Alaska and held in seawater aquaria, that were observed releasing large and small type Hematodinium dinospores within a 60 day observation period.

Month	Sample size	Percentages			Total Percent sporulation
		small spore	large spore	both spores	
			1989		
July/Aug	67	31.3	16.4	4.5	52.2
Sept	84	22.7	3.4	6.8	32.9
Oct	61	0.0	0.0	0.0	0.0
Nov	81	0.0	0.0	0.0	0.0
Dec	28	0.0	0.0	0.0	0.0
Mean		10.8	4.0	2.3	17.0
			1990		
Feb	71	0.0	0.0	0.0	0.0
Mar	113	0.0	0.0	0.0	0.0
Apr	79	0.0	0.0	0.0	0.0
May	100	0.0	1.0	0.0	1.0
June	84	9.5	2.4	0.0	11.9
July	108	3.7	1.8	0.9	6.4
Aug	88	17.1	1.1	0.0	18.2
Sept	56	1.7	5.1	0.0	6.8
Mean		0.3	0.8	0.1	4.9

releasing dinospores could be identified post-mortem by red, sunken intersegmental membranes and slightly opaque, watery hemolymph.

Mortalities were more frequent during the late spring and summer infection and sporulation period (Table 2) and among heavily infected crabs (Table 3). Crabs heavily infected with the BCD parasite were visibly weakened and often also infected with bacteria or the ciliate Paranophrys. Cannibalism during molting, in addition to Hematodinium sporulation and secondary bacterial or ciliate infections, caused crab mortality. As with infection rates and dinospore releases, mortality rates increased steadily until August of both 1989 and 1990. However, total mortality between seasons ($U=33.5$, $0.1<P<0.2$) and between years ($U=20.5$, $P>0.2$) was not significantly different. Total mortality of crabs after the 60 day observation period reached 97% in August, 1989 and 58% in August of 1990 (Table 2). Mortalities generally increased throughout the year in 1990. Mean monthly total mortality for the six months sampled during 1989 equaled 37.4%. Mean monthly mortality from February to September, 1990 equaled 31.9%, with an average for the entire study of 34.0%. Mortality of infected Tanner crabs was 100% in August of 1989, not significantly higher than the 84% mortality observed in 1990.

Mortality rates of infected crabs increased through

Table 2. Possible causes of mortality in Tanner crabs collected from Auke Bay, Alaska and held for 60 days in seawater aquaria.

Month	Sample size	Percent: *Hematodinium* sporulation[a]	Bacterial infection	Ciliate infestation	cannibalism	Total % mortality after 60 days
			1989			
July/Aug	67	61.1	13.4	3.0	0.0	77.5
Sept	84	41.0	9.1	0.0	0.0	50.1
Oct	61	6.5	0.0	0.0	0.0	6.5
Nov	81	0.0	0.0	0.0	0.0	0.0
Dec	28	0.0	0.0	0.0	0.0	0.0
					mean	26.8
			1990			
Feb	71	4.2	0.0	0.0	2.8	7.0
Mar	113	0.9	1.3	0.0	0.0	2.2
Apr	79	0.0	4.0	0.0	0.0	4.0
May	100	1.0	2.4	0.0	0.0	3.4
June	84	17.9	1.0	0.0	0.0	18.9
July	108	9.3	0.0	0.0	0.0	9.3
Aug	88	24.9	3.3	1.1	0.0	29.3
Sept	56	11.9	1.7	0.0	0.0	13.6
					mean	11.0

a)Mortality due to *Hematodinium* sporulation includes those crabs which died during sporulation and heavily infected crabs which were not observed releasing dinospores but had red, sunken intersegmental membranes and opaque, watery hemolymph upon post-mortem examination.

Table 3. Incidence of BCD dinoflagellate infection and percent mortality in Tanner crabs collected from Auke Bay, Alaska before and after 60 day observation periods.

Month	Sample size	Initial # infected crabs	# infected crabs after 60 d	% mortality of infected crabs during 60 d
		1989		
June	195	--	--	--
July/Aug	67	64	0	100
Sept	84	46	1	98
Oct	61	7	4	43
Nov	81	6	6	0
Dec	28	0	a)	
		1990		
Feb	71	8	6	25
Mar	113	23	7	70
Apr	79	0	5	b)
May	100	8	7	13
June	84	18	13	28
July	108	19	10	47
Aug	88	49	8	84
Sept	56	19	8	58

a) No infected crabs were collected during December, 1989 and no crabs were collected in January, 1990.

b) Parasites were not evident in the initial hemolymph samples but were identified in the final hemlymph sample. All crabs in the April sample had recently molted or molted within the 60 day observation period.

August of both 1989 and 1990 (Table 3). Percent mortality equalled about 100% in late July and August and 98% in September of 1989, reaching 84% in August, 1990. These percentages are not significantly different, despite larger sample sizes and fewer infected crabs captured during 1990 ($U=18$, $P>0.2$) (Table 3). Mortality of infected crabs was significantly greater from June to September for both 1989 and 1990 than October through May ($U=32$, $0.02<P<0.05$). (Table 3). Seventy-five percent of uninfected old and new shell female crabs collected in September, 1989 lived until February, 1990 and 45% of these crabs lived until September, 1990; suggesting that crabs experienced little stress during the shorter 60 day holding period (Table 4).

Newly molted crabs were less abundant in samples collected in Auke Bay during 1990 (Table 5). An average of 44.4% of the crabs sampled in 1989 and 46.1% of the crabs sampled in 1990 were newly-molted animals. However, removal of the April, 1990 sample collected from shallower waters (in which 100% of the animals had recently molted) and recalculating the percent new:old shell crabs gives a ratio of 30:70. Significantly more newshell infected and oldshell uninfected crabs occurred during September ($X^2=65.74$, $P<.001$) of 1989 and May ($X^2=6.30$, $.01<P<.025$), July ($X^2=63.79$, $P<.001$), and September ($X^2=16.92$, $P<.001$) 1990, coinciding with the period of increasing incidence of BCD in the population

Table 4. Survival of uninfected old and new-molt Tanner crabs from Auke Bay, Alaska held in seawater aquaria from September, 1989 until September, 1990.

	1989		1990		
Month	Sept	Nov	Feb	May	Sept
Survivals					
#	60	50	45	41	27
%	100	83	75	68	45

Table 5. Comparison of the percent molt stages (newly molted vs. old, encrusted shell) and Bitter Crab Disease prevalence in Tanner crabs sampled from Auke Bay, Alaska from July, 1989 to September, 1990. Percent presence and absence of disease denoted by + and -, respectively.

Month	Sample size	Molt stage Ratio (%new:old)	% Disease Prevalence New-molt (+:-)	old-molt (+:-)	χ^2_c
		1989			
July/Aug	67	78:22	96: 4	93: 7	0.0592
Sept	84	51:49	100: 0	10: 90	65.7401
Oct	61	43:57	8: 92	11: 89	0.0025
Nov	81	21:79	6: 94	8: 92	0.0629
Dec	28	29:71	0:100	0:100	-
Mean % new:old		44.4:55.6			
1990					
Feb	71	52:48	19: 81	6: 94	1.6701
Mar	113	57:43	25: 75	24: 76	0.0248
Apr	79	100:0	6: 94	0: 0	-
May	100	27:73	37: 63	12: 88	6.2956
June	84	-	-	-	-
July	108	16:84	88: 12	4: 96	63.7870
Aug	88	23:77	47: 53	49: 51	0.0476
Sept	56	48:52	72: 28	13: 87	16.9220
Mean % new:old		46.1:53.9			

(Table 5). Assuming equal likelihood of parasitism, shell condition was not independent of infection rate when all months were pooled (X^2_c=112.84, P<.001). Significantly more new-shell and fewer old-shell crabs were infected when all months were combined.

Discussion

In recent years the southeastern Alaska Tanner crab fishery has occurred during the first two weeks of February, when Tanner crabs aggregate in shallower waters prior to molting and mating, and are easier to catch. February is the time of optimum meat yield and condition for most stocks (Imamura and Bothelo, 1989). Unfortunately, unmarketable infected crabs are also captured during this time. Although several processors now collect, cook and grind "sick" crabs thus presumably killing the parasite, culling and disposal of infected crabs on the fishing grounds occurs and the risk of spreading the disease to other areas remains.

A long latency period punctuated by shorter, intense periods of sporulation and host mortality seems typical of Hematodinium parasitism. Hematodinium from the east coast of the United States parasitized blue crab, Callinectes sapidus, in all but the late winter to early spring months (Newman and Johnson, 1975). Likewise, monthly samples from Auke Bay Tanner crabs indicated that parasitism was significantly higher in the summer decreasing to undetectable levels during

mid-winter. Tanner crab heavily infected with BCD parasites were most abundant during August and September in Auke Bay Tanner crabs confirming what had been suggested by a preliminary study undertaken in the Sullivan Island area, southeastern Alaska (Eaton et al., 1991).

The lower estimates of incidence and intensity in Auke Bay during April, 1990 may have been due to atypical sample composition. The April sample, collected from 10 m of water via SCUBA, consisted of smaller, morphologically immature male crabs and primiparous females. All of these younger crabs were newly molted animals which may have been spatially segregated from the larger crabs captured from deeper waters, and possibly less exposed to the dinoflagellate. Although ontogenic spatial segregation has not been documented for southeastern Alaskan C. bairdi, C. opilio in the Sea of Japan undergo ontogenic migrations which result in spatial segregation of adults and juveniles (Kon, 1982). Meyers et al. (1987) reported that BCD infections do not seem size specific in Tanner crabs collected from the Sullivan Island area. No Tanner crabs under 60 mm carapace width were sampled from Auke Bay. Size-specific differential infectivity studies of BCD have not been conducted and are needed.

No interannual variations in disease incidence or intensity occurred between 1989 and 1990. Incidence and intensity of BCD infection was not significantly different in

1989 from 1990 based on Mann-Whitney U non-parametric rank test (Zar, 1974). Tanner crab mortality rates in 1990 at the end of the 60 day observation period were also not significantly lower. While incidence of disease and mortality may not be significantly different for 1989 and 1990, multi-year studies are needed to accurately assess long-term trends in BCD infection rates. The percentage of new-shell crabs sampled in 1990 was not significantly different from 1989 based on Mann-Whitney U test. Several years of sampling are required to determine whether differential mortality of newly molted crabs occurs. Decreasing catch rates and lower average intensity of infection were reported for second-year samples taken near Sullivan Island, Alaska (Meyers et al., 1987).

BCD appears to eventually kill all Tanner crabs which become infected with the parasite. In Auke Bay, mortality rates increased until August and September when sporulation, disease incidence and intensity were highest. All infected crabs held for extended periods beyond 60 days eventually died due to secondary bacterial and ciliate infections or late summer spore release of the BCD dinospores. In general, mortality during the late fall, winter and spring was more likely to be due to bacterial infection then Hematodinium sporulation, which was highest in late summer. Based on this study, the optimal harvest period for Tanner crab in Auke Bay would be November through January when disease incidence and

intensity is relatively low. Research done in upper Lynn Canal concurs with this study, suggesting a harvest period from October to December (Eaton et al., 1991). Managers should plan the fishery for the Lynn Canal area to occur during this time.

Tanner crab mortality due to parasite sporulation and secondary bacterial and ciliate infections may explain the decreasing incidence of BCD seen during the fall and winter. Since the parasite seems highly virulent, killing most infected Tanner crabs, it is unlikely the development of immunity plays any role in decreasing incidence of BCD in the fall and winter months. The source or route of infection during the spring, resulting in an increase in incidence and intensity of BCD, is still unknown. The parasite has been observed exiting the crab via the gills (Love, unpublished data). However, many questions remain. Like the free-living dinoflagellates, do the BCD dinospores serve as disseminatory stages which eventually form overwintering hypnocysts, becoming infective the following spring? Do the dinospores attach to uninfected Tanner crabs and await molting when the unhardened epicuticle could be more easily penetrated much like the sarcodine parasite Paramoeba perniciosa of blue crabs, as hypothesized by Meyers et al. (1990)? Do heavily infected crabs serve as a reservoir of infection, transmitting the disease when cannibalized upon their death?

In previous studies, newly-molted crabs had a significantly higher incidence of BCD infection than old-shell Tanner crabs (Eaton et al., 1991). Significantly more old-molt uninfected crabs occurred in samples taken from Auke Bay during the summer of 1990. BCD-related mortality of new-molt crabs may explain the lower overall ratio of new:old molt crabs in 1990 and the higher incidence of uninfected old-molt crabs. Sample sizes were generally larger in 1990, allowing for more precise estimation of disease incidence and mortality estimates. Continuing study is required to assess the long-term effects of differential mortality on age structure of Tanner crab populations affected with BCD.

Relatively little research has been done on Bitter Crab Disease, with many more questions being asked than answered. Food science studies, development of BCD assays, mortality estimation and parasite distribution studies continue. Disease-related mortality should become a working part of the management of commercially important invertebrates such as Tanner crab. Different harvest periods may need to be established so as to minimize cost of capturing unmarketable, infected crabs and the risk of spreading the disease. Given the value of the Tanner and snow crab fisheries in Alaska and the potential impact this parasite could have on the resource, the results of this research require the attention and action from researchers, management and industry.

CHAPTER III.

SUSCEPTABILITY OF RED KING CRAB (*Paralithodes camtschaticus*) AND DUNGENESS CRAB (*Cancer magister*) TO THE DINOFLAGELLATE PARASITE RESPONSIBLE FOR BITTER CRAB DISEASE IN TANNER CRABS (*Chionoecetes bairdi*)

Introduction

Hematodinium perezi was first described by Chatton and Poisson (1931) in the portunid crabs *Carcinus maenus* and *Portunus depurator* off the Atlantic coast of France. This parasite has also been described in Atlantic rock crabs (*Cancer irroratus*), Jonah crabs (*Cancer borealis*), and lady crabs (*Ovalipes ocellatus*) from the inshore and offshore waters of the mid-Atlantic bight and in blue crabs, *Callinectes sapidus*, from the coastal waters of North Carolina to Florida (MacLean and Ruddell, 1978; Newman and Johnson, 1975). More recently, a parasite similar to *H. perezi* was described from the hemolymph of Tanner crab (*Chionoecetes bairdi*) from southeastern Alaska and Tanner and snow crab (*Chionoecetes opilio*) from the Bering Sea (Meyers et al., 1987; Meyers et al., 1990). The resultant disease has been termed Bitter Crab Disease (BCD).

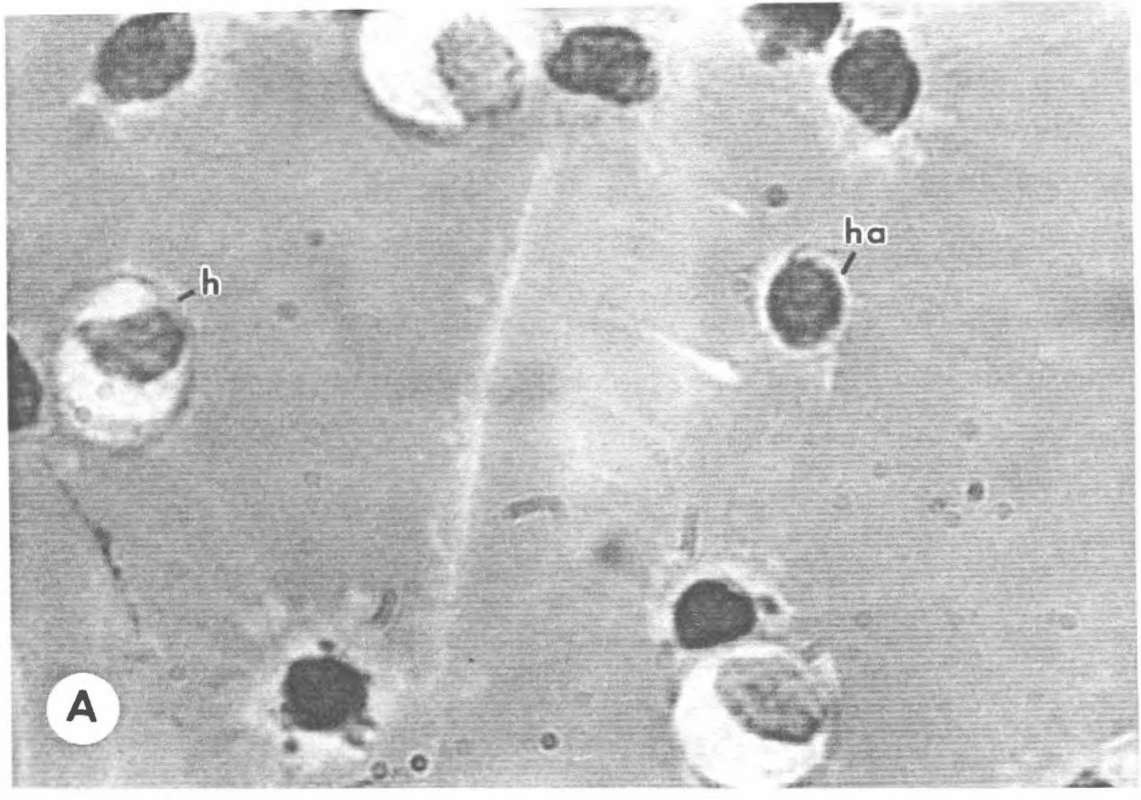
The BCD parasite causes Tanner crab meats to have an astringent, aspirin-like aftertaste during the multiple month residence period within the hosts' hemolymph and tissues.

During July and August the amoeboid form develops into one of two different motile dinospore stages which exit via the gills (Love, unpublished data) thus killing the crab (Eaton et al., 1991). Disease-related losses of revenue and closures of Tanner crab fishing areas have already affected the industry (Imamura and Bothelo, 1989). In Alaska, there is concern that BCD could be transferred to Dungeness and red king crab.

The purpose of this study was to determine the susceptability of the commercially important Dungeness (Cancer magister) and red king crab (Paralithodes camtschaticus) to the BCD parasite. In southeastern Alaska, Dungeness crab fishing supported a $3.8 million industry while the harvest of red and blue king crab was valued at $344,000 during the 1989-1990 season.

Materials and Methods

Red king crabs and Tanner crabs were collected from Auke Bay, Alaska using 2m x 2m conical commercial Tanner crab pots (Figure 3). Dungeness crabs were collected subtidally via SCUBA. Initial hemolymph samples were collected from all crabs, smears were made and stained using Diff-Quik histological stains (Baxter Scientific, Inc). Heavily-infected Tanner crab hemolymph containing amoeboid and spore forms of the parasite was collected from Tanner crab caught during August of 1989. Approximate number of parasites per ml of infected hemolymph was determined with a hemacytometer.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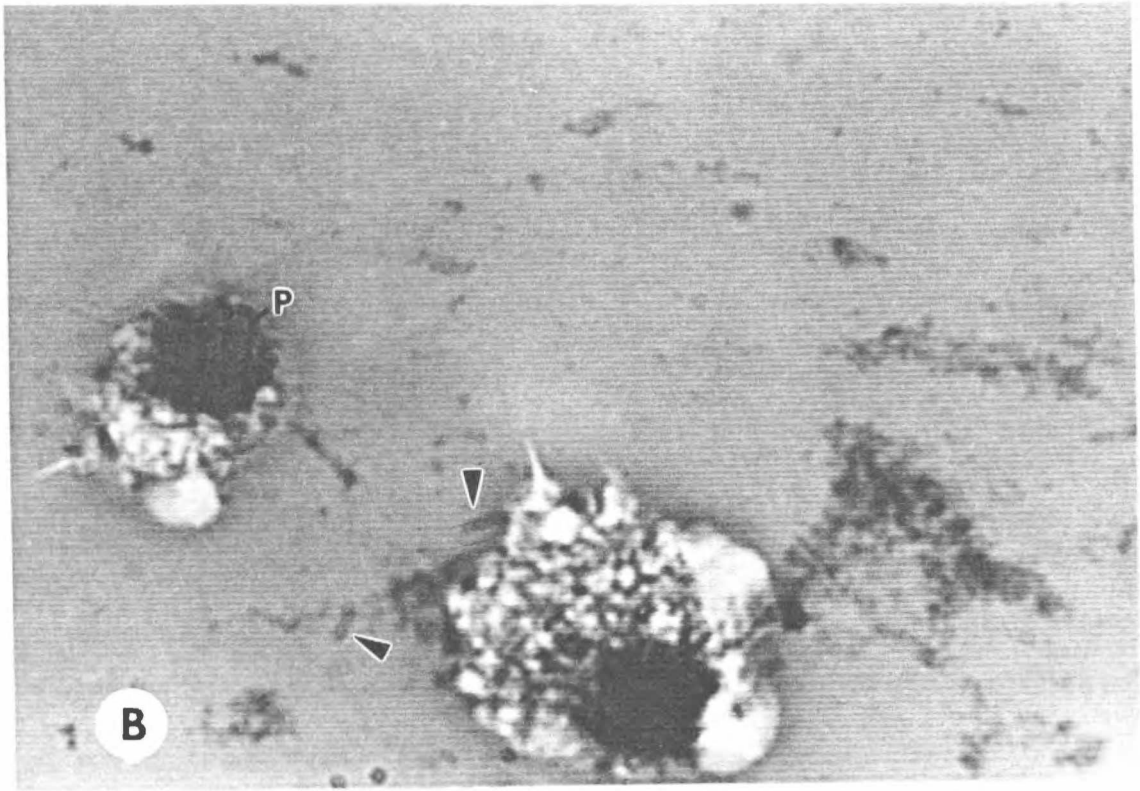
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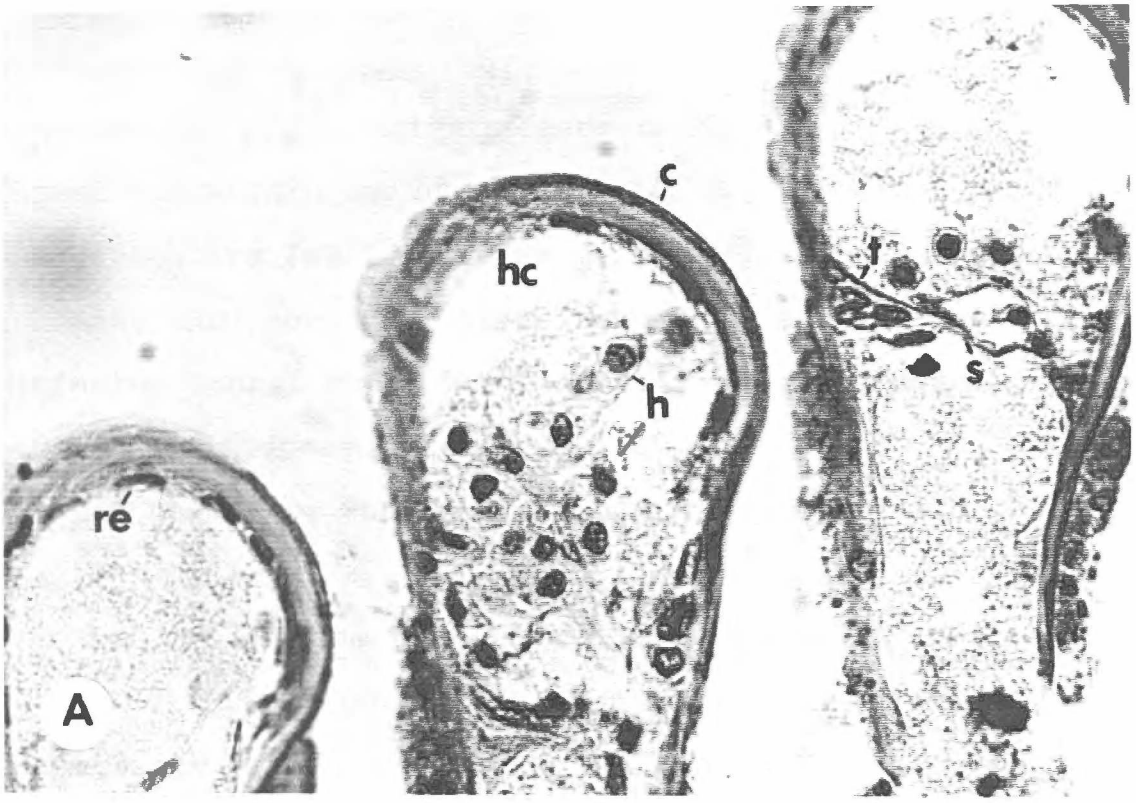
crab hemocytes, later stages of the BCD parasites had indistinct cell membranes, foamy, refractile cytoplasm and an acentric, dinokaryon nucleus (Figure 4). This stage of the parasite had eosinophilic nuclei and could often be seen dividing. Earlier stages of the parasite had proportionately less cytoplasm and basophilic nuclei. Older, vegetative stages of the BCD parasite eventually formed multi-cellular plasmodia which soon lost most of the refractile cytoplasm to the surrounding hemolymph. These plasmodia then metamorphosed into the active dinospores. Eaton et al. (1991) provide microphotographically a complete description of the four amoeboid and two dinospore life-stages of the BCD parasite.

Respiratory and circulatory

The respiratory and circulatory systems were the most heavily impacted by BCD. Uninfected crab gills have easily identifiable cuticle layers underlain by a respiratory epithelium. The hemal channels contain the granular-appearing hemolymph (H&E) and hemocytes (Figure 5). The hemal channels within the lamellae are separated by septae and associated trabecular cells.

Heavily infected crabs have hemal channels within the trabeculae of the gill filled with numerous parasites; septal divisions are less frequent and the respiratory epithelium is often degenerate. These tissues seem more fragile than

Figure 5. Tanner crab gill lamellae of crab from Auke Bay, Alaska. (A) Uninfected Tanner crab gills. (B) Gills from a heavily infected Tanner crab; hematoxylin-eosin, (400X). c, cuticle; re, respiratory epithelium; hc, hemal channels; h, hemocytes; s, septae; t, trabecular cells; dc, degenerate respiratory epithelium; p, BCD parasites.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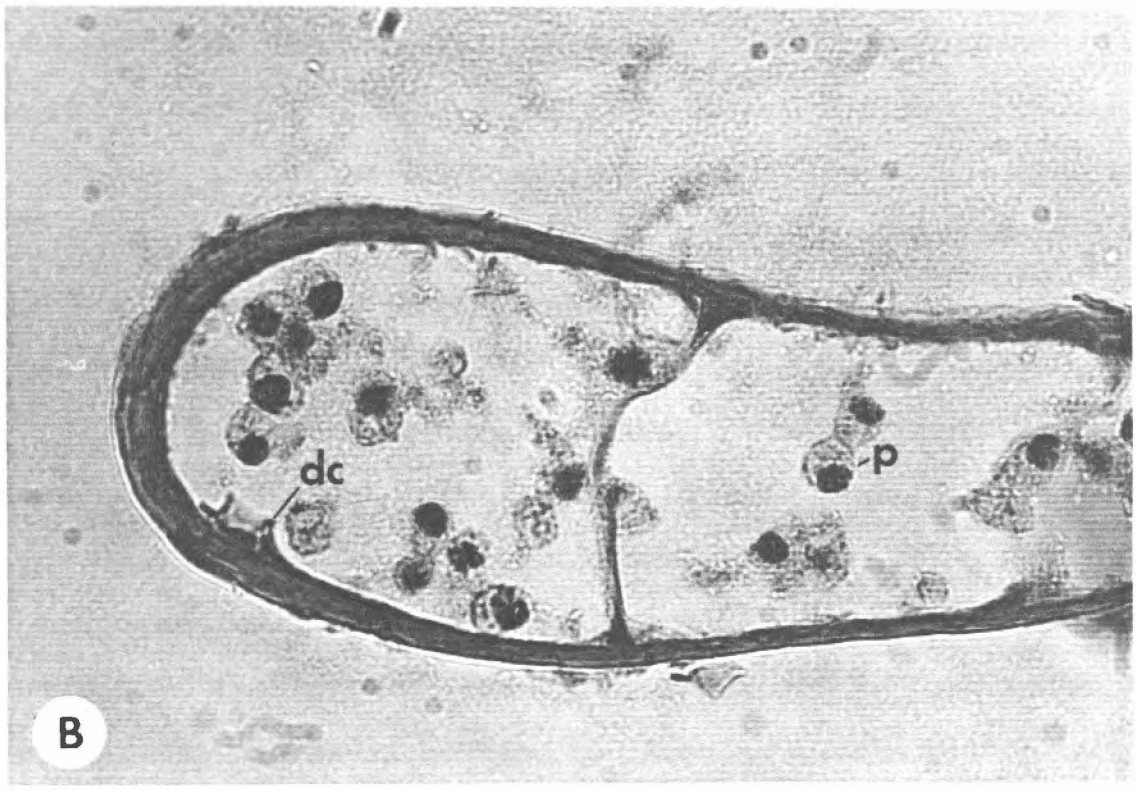
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uninfected gills and are more difficult to section. The hemolymph within the hemal channels appears less granular and hemocytes are few (Figure 5). Breaks in the gill cuticle were present but may have been caused by sectioning. Heavily infected Tanner crabs observed in seawater aquaria, released the BCD dinospores from the gill, the site most likely to retain parasites when crabs were examined post-mortem (Figure 6).

Hemal sinuses throughout the tissues of infected crabs were often occluded with parasites, and cardiac tissue especially was degenerate. Myocardial muscle bundles appearedless numerous in the infected crabs, although a larger sample size is required to verify this observation. The complex network of interconnected sinuses of the myocardium of the heart contained BCD parasites, which remained in the hemal sinuses and blood vessels (Figure 7). BCD parasites did not penetrate into the epicardium.

Digestive system

The digestive system of Tanner crabs is comprised of the muscular esophagus, cardiac and pyloric stomachs, the less muscular mid-, and hind-guts, and the glandular hepatopancreas. The esophagus, cardiac stomach and intestines were all similar in structure, comprised of cuticular and epithelial layers surrounded by circular muscle and connective tissue. Parasite-induced tissue damage seemed to have

Figure 6. Heavily infected Tanner crab collected from Auke Bay, Alaska, releasing cloud of BCD dinospores (dashed line and arrows) from the gill chamber into the water.

Figure 7. Cardiac tissues of Tanner crab sampled from Auke Bay, Alaska. (A) Uninfected crab cardiac tissue. (B) Cardiac tissue from heavily infected crab; hematoxylin-eosin, (400X). h, hemocyte; my, myocardial muscle bundles; dmy, degenerate myocardia; P, BCD parasites(some with distinct dinokaryon nuclei).


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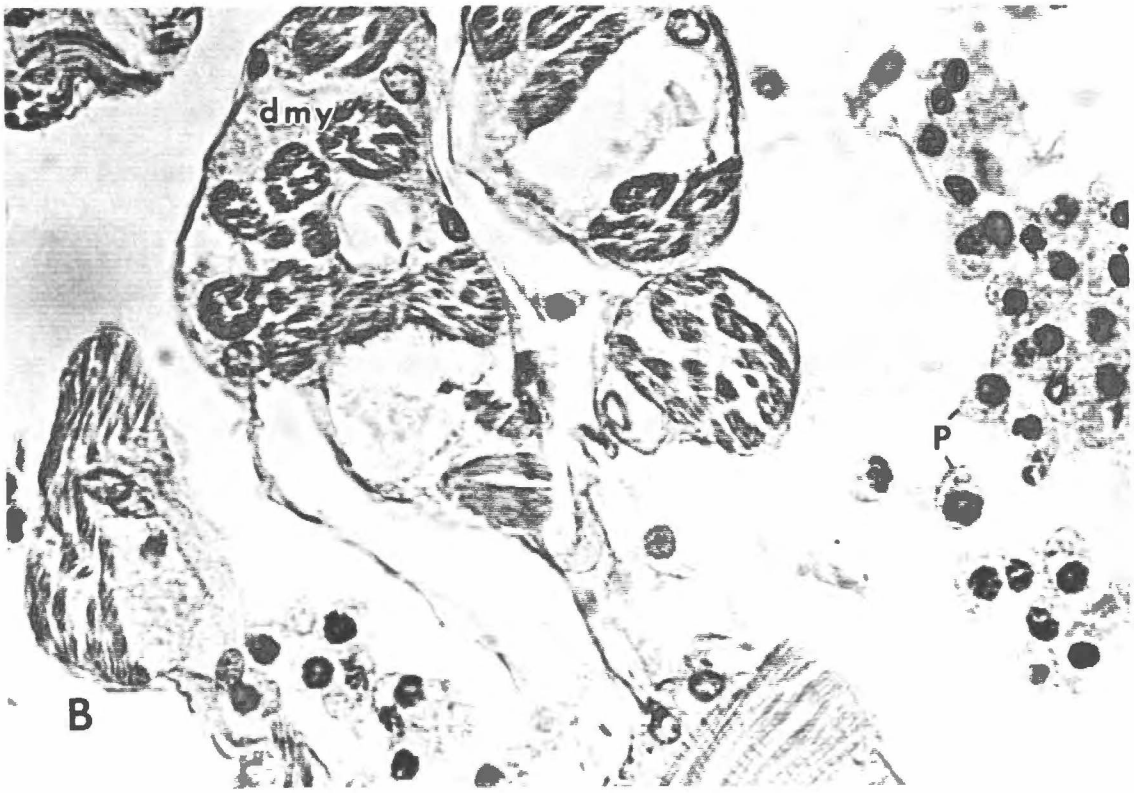
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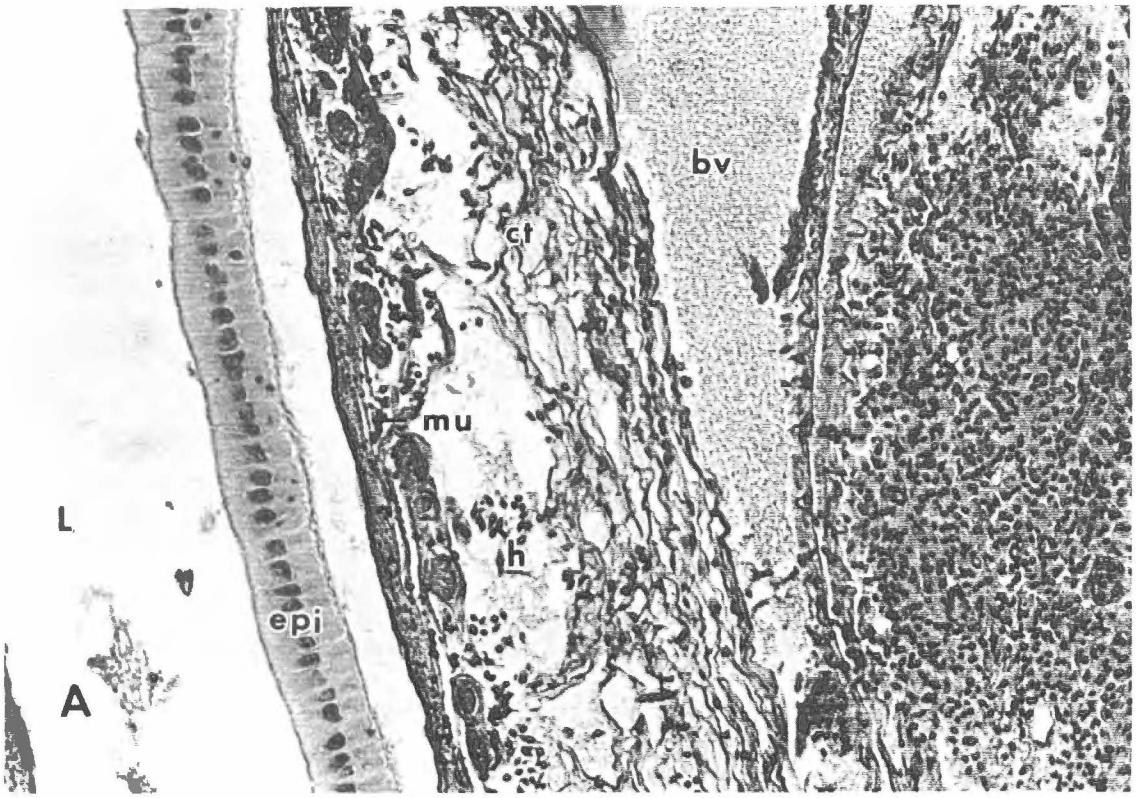
occurred in all these organs. BCD parasites were found in the hemal sinuses adjacent to the epithelial layers of the esophagus, cardiac stomach and gut, causing tissue damage to localized areas of the spongy connective tissue (Figure 8). The hepatopancreas was the most heavily damaged digestive organ. The hepatopancreatic tubules of lethargic, non-feeding, infected crabs had lumens which were ragged in appearance, glandular epithelium which had atrophied and numerous parasites within the interstitial capillaries, arteries and connective tissues (Figure 9). In healthy crabs, the numerous hepatopancreatic tubules had distinct star-shaped lumens and vacuolate glandular epithelium.

Epithelial and glandular

Antennal glands and endodermis from infected and uninfected Tanner crabs were observed via histological sectioning. BCD parasites were common in the blood vessels and hemal sinuses of these tissues and hemocytes were rare in heavily infected crabs. The hemal sinuses of the antennal glands of uninfected crabs were replete with hemocytes. Few hemocytes occurred in the blood vessels of antennal glands from infected Tanner crabs and interstitial tissues were often destroyed. A few parasites were observed within the lumens of the glandular epithelium and between the layers of epithelium (Figure 10).

Macroscopically, the exterior surface of the subcuticular

Figure 8. Mid-gut of Tanner crab from Auke Bay, Alaska. (A) Mid-gut from uninfected crab. (B) Mid-gut from infected Tanner crab; hematoxylin-eosin, (400X). L, lumen of mid-gut; epi, epithelium of mid-gut; mu, longitudinal muscles of mid-gut; h, hemocytes; ct, connective tissues; bv, blood vessel; bc, basal cell; bm, basement membrane; tgl, tegmental gland; pm, peritrophic membrane; p, BCD parasites.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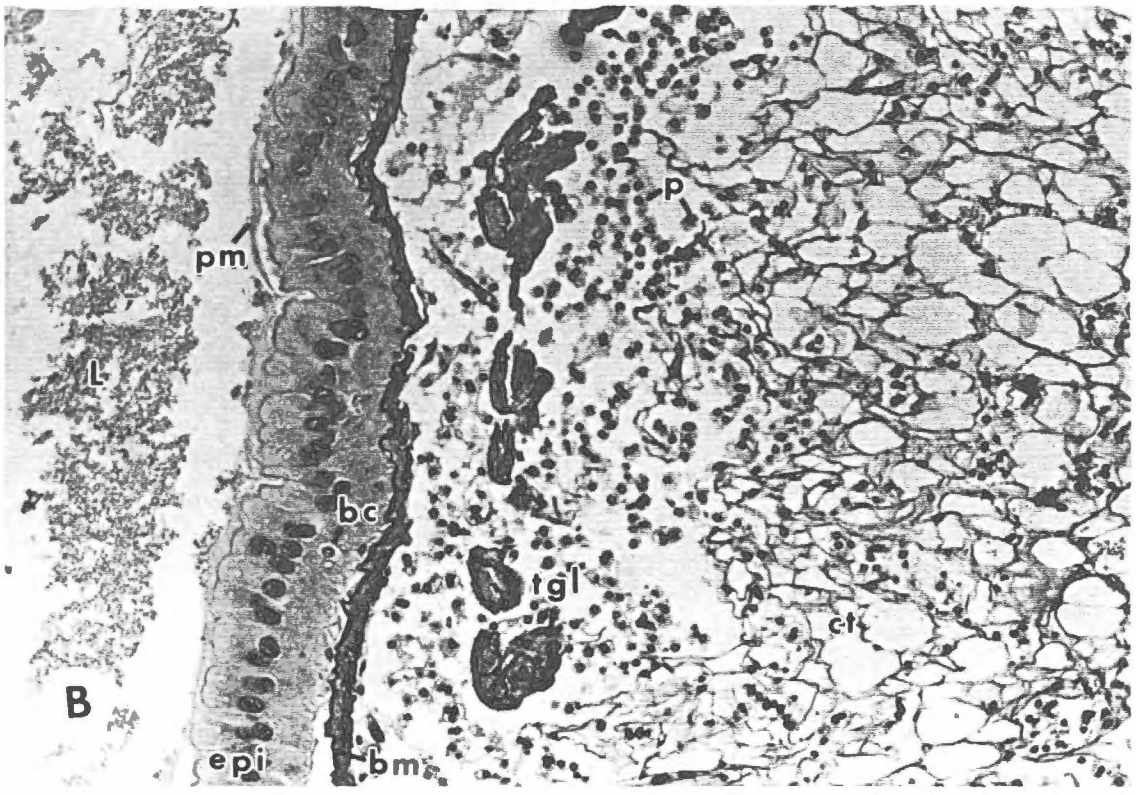
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Figure 9. Hepatopancreatic tubules of Tanner crab taken from Auke Bay, Alaska. (A) Uninfected crab with hepatopancreatic tublues having distinct star-shaped lumens. (B) Infected Tanner crab with degenerate hepatopancreatic tubules having ragged lumens; hematoxylin-eosin, (400X). L, lumens; v, vacuole of B-cell; hs, hemal sinus; he, hepatopancreatic epithelium, i, interstitial tissues; dL, degenerate lumen; P, BCD parasites.

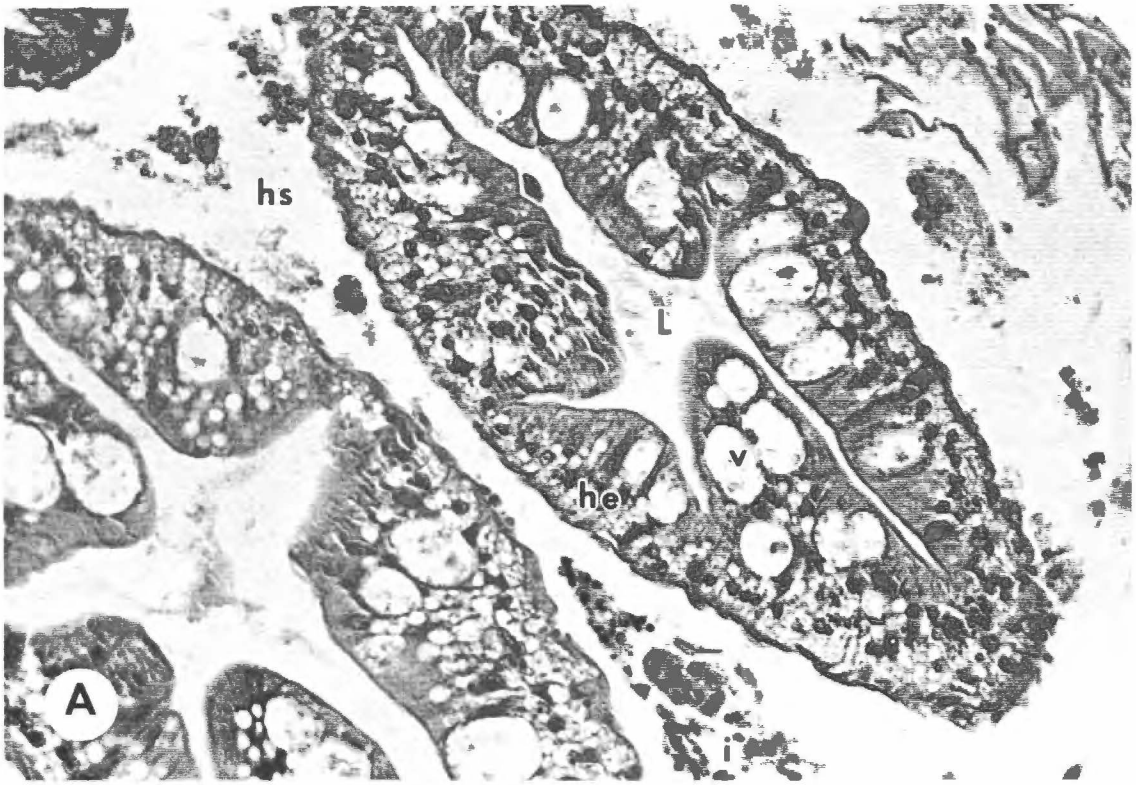
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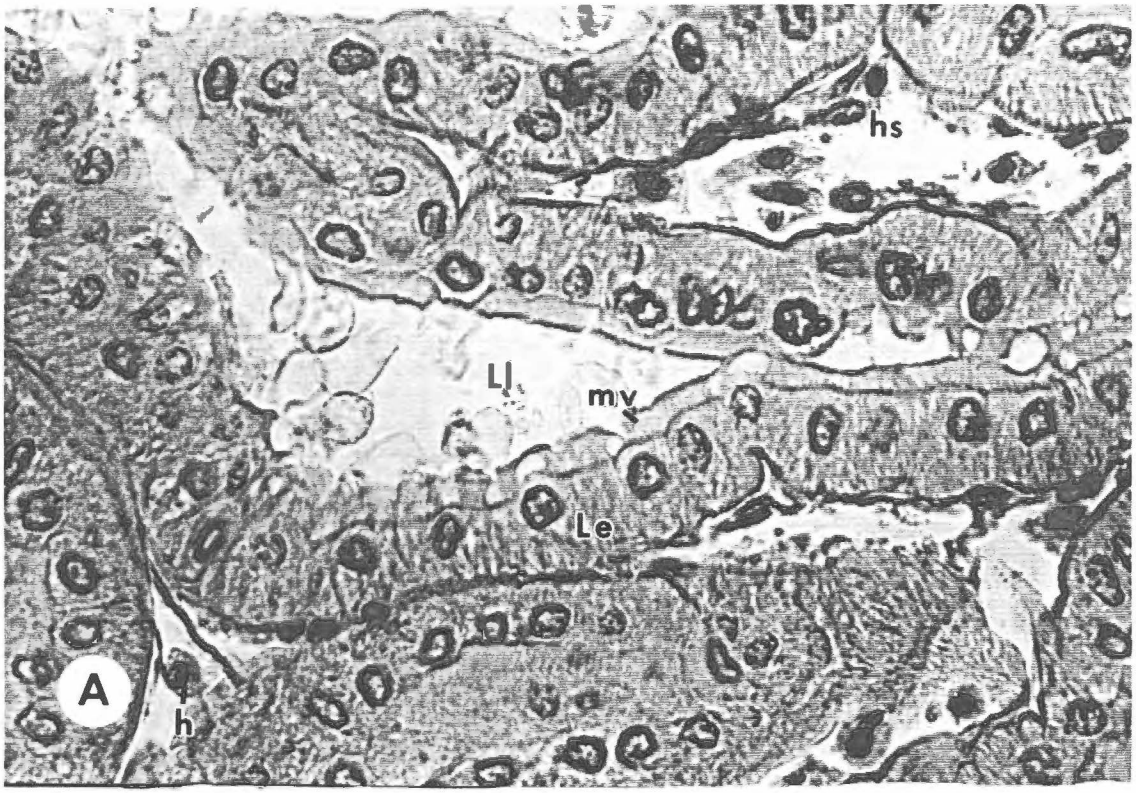
Figure 10. Antennal glands of Tanner crab from Auke Bay, Alaska. (A) Uninfected antennal glands of Tanner crab. (B) Infected antennal glands; hematoxylin-eosin, (400X). Li, lumen of labyrinth; Le,epithelium of labyrinth; Cl, lumen of coelomsac; mv, microvilli; hs, hemal sinus; h, hemocyte; p, BCD parsites.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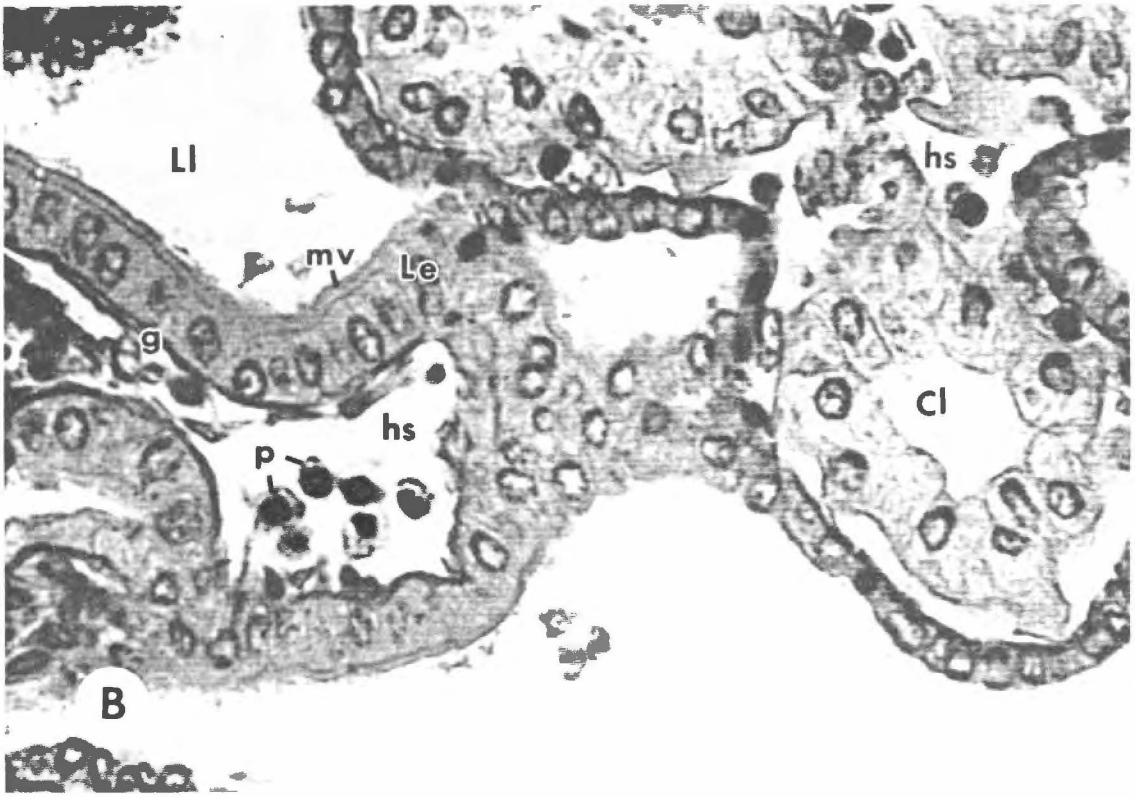
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endodermis from heavily infected crabs was orange in color rather than the normal red. The orange color permeated the tissue. The pink color of the exoskeleton of infected crabs may be caused by the leaching of melanins and destruction of melanocytes (Meyers et al., 1987). Uninfected endodermal tissue was darker red in color, and crabs had an exoskeleton more brown in color. Parasites were found within the luminal spaces adjacent the endothelium as well as exterior to the newly forming cuticle, causing some disruption of the hemal sinuses and connective tissues. (Figure 11).

Muscular, nervous and reproductive tissues

Meats from the walking legs of lethargic, infected crabs were one-half to one-third the size of uninfected Tanner crab leg-meat. Microscopically, muscle cells and the surrounding connective-tissue capsule from parasitized crabs appeared intact, with BCD parasites between the muscle bundles and within the blood vessels (Figure 12).

The supraesophageal and thoracic ganglia were not directly parasitized by the BCD parasites. Hemal channels and blood vessels supplying these nerves did carry the parasites. Nervous tissue from infected crabs appeared similar to that of uninfected crabs.

Reproductive tissues had parasites imbedded among the interstitial cells far from hemal sinuses and blood vessels. Although spermatophores and eggs were not destroyed by this

Figure 11. Endocuticle and endothelium of Tanner crab from Auke Bay, Alaska. (A) Uninfected tissues. (B) Infected tissues; hematoxylin-eosin, (400X). c, cuticle; epi, epithelium; hs, hemal sinus; h, hemocyte; ct, connective tissue; tgl, tegmental gland; p, BCD parasite.

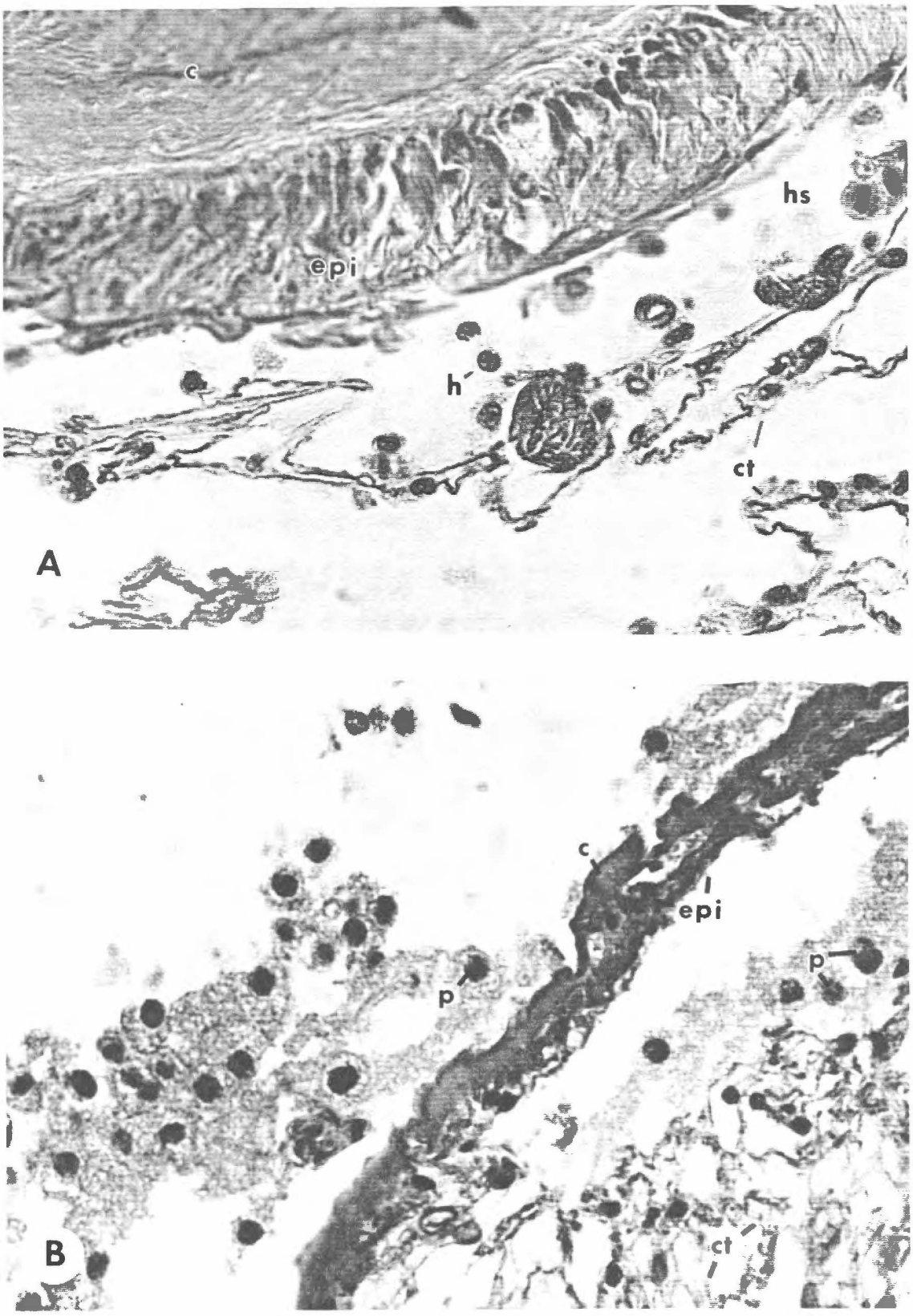
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Figure 12. Reproductive tissues of Tanner crab from Auke Bay, Alaska. (A) Ovary from lightly infected Tanner crab. No BCD parasites are shown. (B) Testis from heavily infected Tanner crab; hematoxylin-eosin, (400X). Oc, Oocyte; N, nucleus; Nu, nucleolus; ac, accessory cell; i, interstitial tissues; sp, spermatids; P, BCD parasites.


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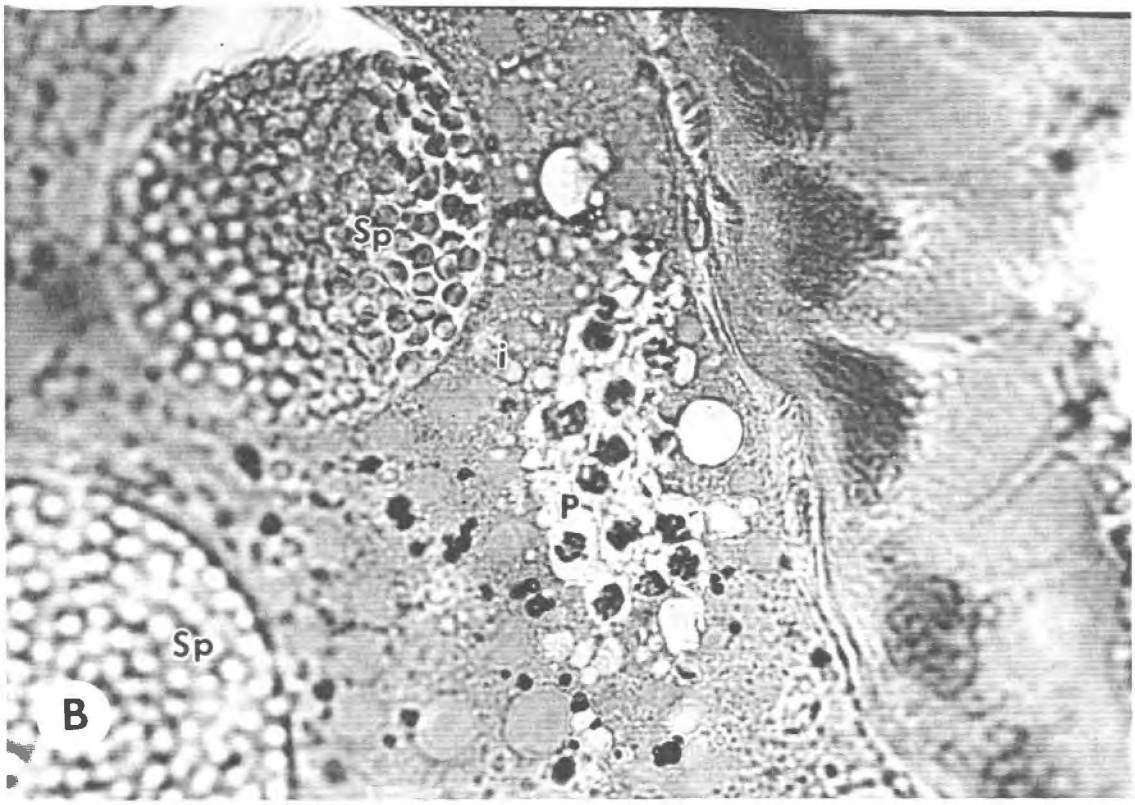
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encystment, subsequent metamorphosis and dinospore release into the hemolymph must be damaging (Figure 13). BCD parasties are not present in the picture of the ovary. Reproductive tissues from uninfected crabs were not sampled.

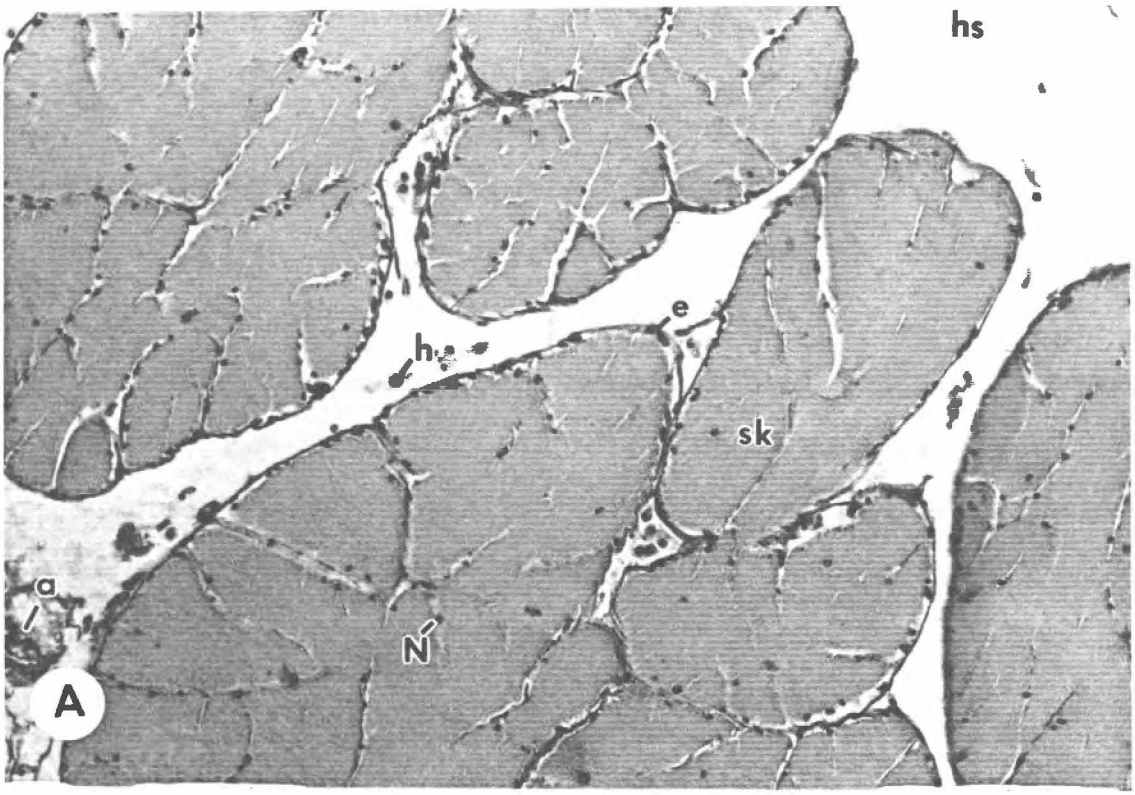
Discussion

Parasite challenges

The route of transmission of BCD in the wild remains undefined, although the portal of exit appears to be via the gills. Waterborne dinospore challenges did not result in disease in old molt Tanner crab and have not been successful with newly molted crabs currently held at the NMFS Auke Bay Lab. However, none of these crabs have been held through the molt. Meyers et al. (1987) suggested that BCD dinospores released in late summer attach to the carapace of new hosts to await the spring molt when the cuticle may be more easily penetrated. This method of transmission is similar to that suggested for Paramoeba pernisciosa which parasitizes blue crab, Callinectes sapidus (Couch, 1983). BCD parasite waterborne challenges of pre-molt Tanner crab are needed to determine if this route of transmission is possible.

In the laboratory, BCD can be transferred to new hosts via injection into the hemolymph. Vegetative cells have consistently caused disease following injection (Meyers et al., 1987; Eaton et al., 1991). In the current study,

Figure 13. Skeletal and cardiac muscle tissue of Tanner crab from Auke Bay, Alaska. (A) Skeletal muscle from uninfected crab. (B) Cardiac tissue from heavily infected crab; hematoxylin-eosin, (100X). hs, hemal sinus; h, hemocyte; sk, skeletal muscle bundle; e, epimysium; N, sarcoplasmal nucleus; a, arteriole; Epi-, epicardium; My-, myocardium; sct, spongy connective tissue; fi, myofibril myocardial muscle bundle; p, BCD parasites.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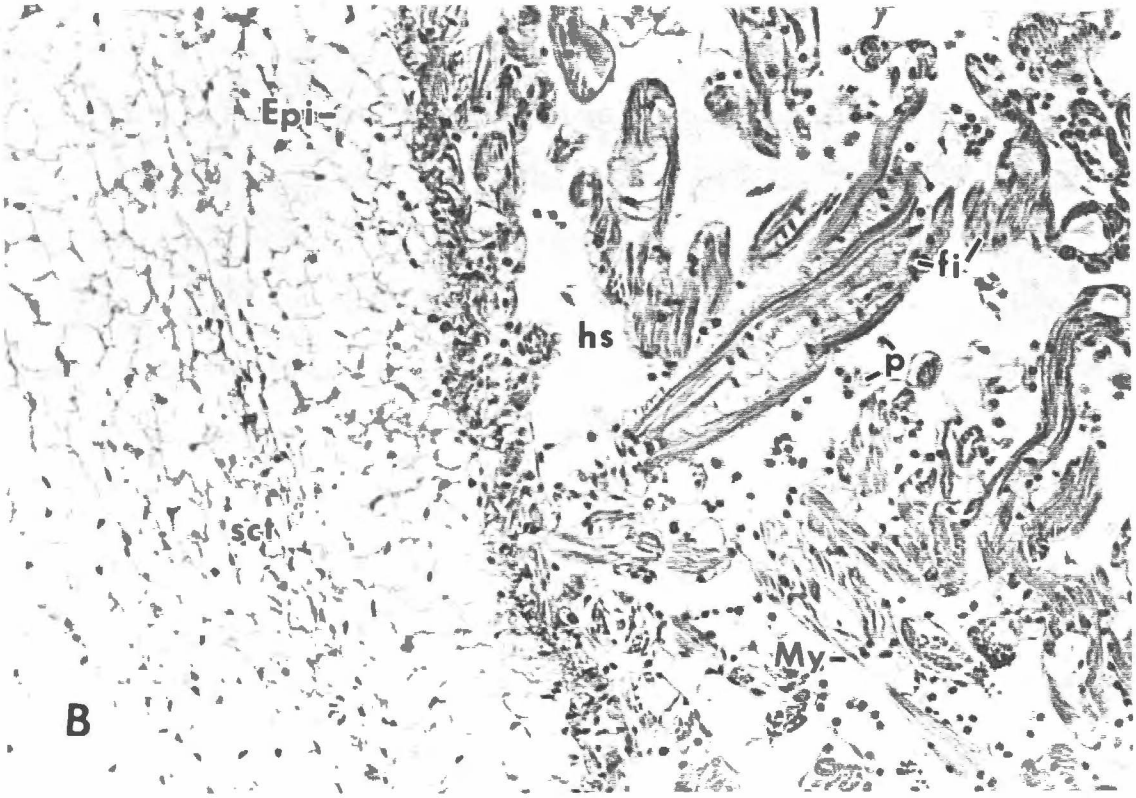
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intensity of BCD infection and sporulation periods among the crabs challenged with BCD vegetative cells mimicked that observed in infected Tanner crabs sampled concurrently from Auke Bay.

Some of the crabs that were injected with BCD dinospores developed the disease. Three months post-injection, BCD dinospores caused disease in two Tanner crabs from each group of 14 challenged with large dinospores and a 50:50 mixture of large and small dinospores, respectively (Table 2). The remaining crabs challenged with BCD dinospores did not develop the disease and eventually died from secondary bacterial infections. All vegetative-challenged crabs eventually expressed the disease.

The purpose of the BCD dinospore remains undefined. Like some of the free-living dinoflagellates, the BCD dinospores may aid in dissemination of the parasite and may develop into a resting stage, which later becomes infective. Dinospores have survived in sterile seawater for as long as 73 days post-inoculum (Meyers et al., 1987).

Estimates of the mass of DNA in the vegetative and spore forms of the BCD parasite were similar for all, suggesting a similarity in ploidy (Eaton et al., 1991). This could mean that different-sized dinospores are not separate sexes or separate gametes. In addition, both Eaton et al. (1991) and Love et al. (1992) found that an individual crab can harbor

both of the 2 spore types, in contrast to the findings of Meyers et al. (1987). Thus, it may be that there are two different but related species of BCD dinoflagellates or two forms of the same species. An extensively studied free-living dinoflagellate, Crypthecodinium cohnii, produces morphologically similar but genetically discrete syngens due to reproductive isolation (Beam and Hines, 1984). If the BCD dinoflagellates reproduce (plasmodial stage) within an individual crab host, the chances for genetic as well as morphological variation in the dinospores may be enhaced. Genetic comparisons of the micro- and macro-spores may help bring about a better understanding of the role the dinospores in the epizootiology of BCD.

BCD parasites may also be transmitted to new hosts when weakened, infected crabs are cannibalized. Preliminary results of a study initiated in the fall of 1990 at NMFS Auke Bay Lab indicate that food-borne challenges may result in disease (Love and Moles, unpublished). Several months of study are still required. This may mean that the vegetative stages of the parasite infect the new host via the digestive system. If the disease can be transmitted by cannibalism, the impact of disposal of infected crab on uninfected crab stocks could be damaging and more infected crab should be retained by the fishermen and delivered to the processors for grinding, cooking and disposal. Additional research will determine if

lightly infected crabs can be marketed or otherwise utilized.

Histological observations

Given the open circulatory system found in decapods, one might expect that the various tissues thus exposed to the activity, metabolites and irritations of the BCD parasites would be degenerate or destroyed. Throughout the organs of the Tanner crab, tissue destruction seemed largely confined to the hemal sinuses, blood vessels and surrounding connective tissues. As the disease progressed, fewer hemocytes were observed in subsequent hemolymph samples, potentially leaving the crab immunosuppressed and vulnerable to any pathogenic bacteria able to penetrate the cuticle. Non-self recognition of antigens is observed among hemocytes found within the hemolymph of blue crabs, C. sapidus. Throughout the long infection period, hemocytes were not observed aggregating around or attempting to phagositize the BCD parasites indicating that they were not considered foreign. In heavily infected crabs, the gills became severely occluded with parasites limiting oxygen exchange and the respiratory epithelium began to degenerate. Lethargic, infected crabs did not feed. These same crabs had previously fed enthusiastically.

In addition to the bitter taste of infected crabs, leg meats from heavily infected crabs were atrophied, making potential harvest from the wild less desirable. In general,

the condition of the crab steadily deteriorated as the disease progressed.

Parasites in heavily infected Tanner crabs were observed histologically within the lumen of the esophagus, digestive glands and within the gill. Presence of parasites within the lumen of the antennal glands may indicate another potential site of release. However, waterborne challenges of Tanner crabs have to date been unsuccessful. If healthy crabs can be infected by cannibalizing infected crabs, cannibalization might be an important factor in the spread of disease, in addition to whatever role the BCD dinospores play. Additional research addressing this question is needed. Future histological sampling should include mandibular organs, Y-organs and the hemopoietic tissues from the dorsal surface of the cardiac stomach.

CONCLUSIONS

1) The seasonal incidence and intensity of Bitter Crab Disease (BCD) in Auke Bay Tanner crabs decreases to undetectable levels by December and January.

a) BCD is a fatal disease, and most infected crabs eventually die. This mortality should be taken into account when managing the Tanner crab fisheries.

b) The commercial harvest of Tanner crabs should be limited to the period from November to February when infection rates are lowest.

2) A sample of ten Dungeness and red king crab were not susceptible to BCD.

3) Injected amoeboid stages of the BCD parasite will consistently cause disease in old-molt Tanner crab; large dinospores and a 50:50 mixture of large and small spores can cause disease when injected into the crabs hemocoel.

4) Waterborne challenges of old-molt Tanner crabs with BCD dinospores to date have not caused disease.

5) BCD parasites throughout the organs of the Tanner crab seem to be confined to the hemal and blood vessels.

a) In addition to the hemal sinuses and blood vessels, BCD parasites also occurred interstitially in the reproductive organs.

b) Few hemocytes occur in heavily infected crabs, indicating that these tissues have been almost completely destroyed.

RECOMMENDATIONS

1) When sampling to determine disease incidence and intensity, a target of 100 crabs collected from randomly placed pots should be the goal. Hemolymph samples should be taken as soon as the crabs reach the lab to avoid missing infected crabs that eventually die.

2) Multi-year studies of the incidence and intensity of BCD are needed to determine the interannual variability of this disease in affected crab populations, the associated mortality and effects on age structure.

3) Histological examination of Tanner crabs having different levels of BCD infection is needed. The organs that should be compared include: antennal glands, mandibular organs, Y-organ, hepatopancreas, intestine, cardiac and pyloric stomachs, hemopoietic tissue, cardiac and skeletal muscle, endodermis, and gill.

4) Species susceptability studies involving red, blue and brown king crab and Dungeness crabs are needed. Sample sizes of 20 injected and 20 control crabs are suggested for statistical analysis allowing for mortality losses.

5) Life history studies should continue to attempt to determine the route of transmission.

6) Full-scale food science studies are needed to determine palatability and consumer acceptability and the compound responsible for the bitter flavor needs to be identified.

7) Development of a BCD immunoassay or chemical test would enable processors and possibly fishermen to test for the disease on the fishing grounds.

LITERATURE CITED

Beam, C.A., and M. Himes, 1984. Dinoflagellate Genetics. 3:263-298. in Spector, D.L. 1984. Dinoflagellates. Academic Press, New York, New York.

Chatton, E., and R. Poisson. 1931. Sur l'existence, dans le sang des crabs, de peridiniens parasites Hematodinium perezi n.g., n. sp. (Syndinidae). C.r. Seanc. Soc. Biol. 105:553-557.

Couch, J.A. 1983. Diseases caused by protozoa. 79-108. in Provenzano, Jr., A.J. (editor) 1983. The biology of Crustacea: Pathobiology. Academic press. New York, New York.

Dodge, J.D. 1984. Dinoflagellate Taxonomy. 2:17-42. in Spector, D.L. 1984. Dinoflagellates. Academic Press. New York, New York.

Eaton, W.E., D.C. Love, C. Botelho, T.R. Meyers, K. Imamura, T. Koeneman. 1991. Preliminary results on the seasonality and life cycle of the parasitic dinoflagelle causing Bitter crab disease in Alaskan Tanner Crabs (Chionoecetes bairdi). Journal of Invertebrate Pathology. 57:426-434.

Imamura, K.K., C.A. Botelho. 1989. Report to the Board of Fisheries Southeast Alaska(Statistical Area A) and Yakutat(Statistical Area D) Tanner crab. 1988/1989: 3.0-3.20.

Jepps, M. W. 1936. On the protozoan parasites of Calanus finmarchicus in the Clyde Sea area. Q.J. Microsc. Sci. 79:589-662.

Johnson, P.T. 1986. Parasites of benthic amphipods: dinoflagellate (Duboscquodinida:Syndinidae). United States Fisheries Bulletin. 84:605-615.

Johnson, P.T. 1980. Histology of the blue crab, Callinectes sapidus: A model for the Decapoda. Praeger Scientific Publishers, Inc., New York, New York. 391 pages.

Kon, T. 1982. On the planktonic larval life of the Zuwai crab, Chionoecetes opilio, occurring along the coasts of the Central Japan Sea. Proceedings of the International Symposium on the Genus Chionoecetes. Alaska Sea Grant Report No. 82-10, December 1982.

Love, D., S. Rice, A. Moles and W. Eaton. 1992. Seasonal mortality due to Bitter Crab Disease and the prevalence and intensity of infection with the Bitter Crab dinoflagellate in Alaskan Tanner crabs from Auke Bay, Alaska. Pending publication.

Love, D. and A. Moles. Transmission studies of Bitter Crab Disease (BCD), waterborne, food and injection challenges. Pending publication.

Manier, J.F., A. Fize and H. Grizel. 1971. Syndinium gammari n. sp. peridnien Duboscquodinida Syndinidae, parasite de Gammarus locusta (Lin.) Crustace Amphipode. Protistologica. 7:213-219.

Maclean, S.A., and C.L. Ruddell. 1978. Three new crustacean hosts for the parasitic dinoflagellate Hematodinium perezi (Dinoflagellata:Syndinidae). Journal of Parasitology 64:158-160.

Meyers, T.R., Koeneman, T.M., Botelho, C., Short, S. 1987. Bitter crab disease: a fatal dinoflagellate infection and marketing problem for Alaskan Tanner crabs Chionoecetes bairdi. Diseases of Aquatic Organisms. 3:195-216.

Meyers, T.R., C. Botelho, T.M. Koeneman and K. Imamura 1990. The Distribution of the bitter crab dinoflagellate syndrome in southeastern Alaskan Tanner Crabs Chionoecetes bairdi. Diseases of Aquatic Organisms 9:37-43.

Newman, N.W., Johnson, C.A. 1975. A disease of blue crabs (Callinectes sapidus) caused by a parasitic dinoflagellate, Hematodinium spp. Journal of Parasitology. 61:554-557.

O'Bannon, B.K. (ed). 1990. Fisheries of the United States, 1989. Current Fisheries Statistics No. 8900. NOAA/NMFS. Silver Spring, MD. 52-70.

Ossiander, F.J. and G. Wedermeyer. 1970. Computer program for sample sizes required to determine disease incidence in fish populations. Journal of the Fisheries Research Board of Canada. 30:1383-1384.

Wickham, D.E., and A. Kuris. 1989. Brood mortality estimation and role of symbionts in egg mortality of the Tanner crab Chionoecetes bairdi in Alaskan waters. 397-402 in Proceedings of the International Symposium on King and Tanner crabs. Alaska Sea Grant College Program Report 90-04.

Zar, J.H. 1974. Biostatistical Analysis. Prentice-Hall, Inc., Englewood Cliffs, N.J. 59-68.

APPENDIX A

Table 8a. June, 1989 samples for BCD intensity from Tanner crabs sampled from Auke Bay, Alaska. IOF=intensity of infection.

Crab #	IOF	Crab #	IOF	Crab #	IOF
101	1	138	1	174	0
102	2	139	0	175	2
104	0	140	2	176	1
105	0	141	0	177	2
106	0	142	0	178	0
107	0	143	1	179	1
108	2	144	3	180	3
109	0	145	1	181	3
110	0	146	2	182	0
111	1	147	1	183	0
112	1	148	2	184	1
113	0	149	1	185	1
114	1	150	1	186	2
115	1	151	2	187	0
116	2	152	2	188	1
117	2	153	2	189	2
118	3	154	2	190	1
119	0	155	0	191	3
120	1	156	3	192	0
121	1	157	1	193	1
122	2	158	1	194	4
123	1	159	0	195	0
124	1	160	1	196	2
125	0	161	0	197	1
126	0	162	1	198	2
127	0	163	1	199	3
128	1	164	0	200	3
129	1	165	3	201	2
130	1	166	1	202	1
131	1	167	1	203	2
132	1	168	0	204	3
133	3	169	2	205	3
134	3	170	1	206	1
135	3	171	2	207	1
136	0	172	0	208	1
137	1	173	1	209	0
210	0	246	3	283	0
211	1	247	1	284	0

Table 8a, continued.

Crab #	IOF	Crab #	IOF	Crab #	IOF
212	0	248	3	285	1
213	2	249	3	286	4
214	2	250	0	287	2
215	0	251	1	288	2
216	2	252	1	290	1
217	2	253	1	292	0
218	1	254	0	293	1
219	2	255	0	294	1
220	1	256	3	295	1
221	1	257	0	296	2
222	3	258	2	297	2
223	0	259	2	298	1
224	1	260	1	299	3
225	0	261	1		
226	0	262	0		
227	1	263	1		
228	0	264	1		
229	0	265	2		
230	1	266	1		
231	4	267	0		
232	2	268	1		
233	4	269	2		
234	0	270	2		
235	2	271	1		
236	1	272	2		
237	0	273	2		
238	2	274	3		
239	2	275	3		
240	3	276	3		
241	1	278	3		
242	3	279	2		
243	0	280	1		
244	0	281	2		
245	2	282	1		

Table 8b. Combined July and August sample, 1989 for BCD intensity before and after 60-day observation period, date and cause of mortality, sporulation and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	spore type released	shell type
II	4		8/8	bacteria	l	1
19	4		8/6		l	1
39	4		8/6	bacteria	l	1
30	4		8/6	veg,bact	s,l	1
IX	4		8/15	bacteria		1
51	4	4	??		s	1
15	4		??	??	sporu-?	1
49	4		8/14	bacteria	l	1
XI	4	4	8/2	veg	s	1
VIII	4		8/4	veg	l	1
50	4		8/17	veg,bact		1
22	4		8/14	bacteria	l	1
20	4		8/9	bacteria	sporu-?	1
41	4	4	8/22		s	1
36	4		8/5	veg	??	1
18	4		8/3		l	1
38	4		8/14	bacteria	l	1
563	4		8/17	motveg,b	sporu-?	1
530	4		8/7	veg	l	1
23	4		8/6	veg, bact		1
29	4		8/9	bacteria	l	1
V	4		??	??	?	1
24	4	4	8/6	veg,bact	s	1
26	4	4	8/23	veg	s	1
544	4		8/7		l	1
63	4		8/7		l	2
54	4		8/15		motile v	2
IV	4		8/15	bacteria	sporu-?	1
61	4	4	8/17	bacteria	s	2
59	4		8/3	Paranophrys		2
57	4		8/22		l	2
67	4		9/5	bacteria		2
VI	4	4	8/25	bacteria	s	1
I	4		8/15	bacteria	sporu-?	1
562	3		8/7	bacteria	l,s	1
46	3		8/4		l	1
595	3		8/8	3+	histosam	1
40	3		8/9	bacteria	l	1
66	3		8/6		l,s	2

Table 8b, Continued.

Crab #	intensity of infection initial	final	date of death	cause of death	spore type released	shell type
27	3	4	8/18		s	1
537	3		8/8	1+ histosam		1
65	3	4	8/26		s	2
44	2		8/14	bacteria	l	1
31	2		8/14	2+ DNAsam		1
597	2		8/8	2-3+ histosam		1
52	2		8/29		l	1
21	2		8/6	bacteria		1
VII	2		8/8	2-3+,bact histosam		1
17	2		8/14	bacteria	l	1
55	2		8/17	veg,bact		2
64	2	4	9/4	bacteria	s	2
522	2		8/4	veg,bact	l	1
III	1		7/31	1+, bact		1
584	1		8/8	1+redhemohistosam		1
575	1		8/8	4+ histosam		1
32	1	4	8/15		s	1
X	1		8/8	3+ histosam		1
48	1		8/18	bacteria	l	1
34	1		8/14	3+ DNAsam		1
45	1		8/6	motveg,b sporu-?		1
53	1		??	?	??	2
56	1		8/17	bacteria		2
62	1		9/15	bact, Paranophyrs		2
58	1		8/23		rnd l	2
37	0		8/6	bacteria		1
43	0		10/1	released	0	1
60	0		10/1	released	0	2

Table 8c. September sample, 1989 for BCD intensity before and after 60-day observation period, date and cause of mortality, sporulation and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	spore type released	shell type
283	0	0	10/18	set aside		2
280	0	0	"	" as		2
289	0	0	"	" control		2
271	0	0	"	" crabs		2
287	0	0	"	" " "		2
255	0	0	"	" " "		2
268	0	0	"	" " "		2
915	0	0	"	" " "		2
272	0	0	11/15	0;released		2
257	0	0	10/18	controls		2
293	0	0	"	" " "		2
252	0	0	"	" " "		2
861	0	0	"	" " "		2
260	0	0	"	" " "		2
286	0	0	"	" " "		2
279	0	0	"	" " "		2
258	0	0	"	" " "		2
264	0	0	"	" " "		2
265	0	0	"	" " "		2
292	0	0	"	" " "		2
250	0	0	11/15	0;released		2
299	0	0	10/18	controls		2
278	0	0	11/15	0;released		2
274	0	0	"	" "		2
267	0	0	10/18	controls		2
297	0	0	"	" " "		2
276	0	0	"	" " "		2
269	0	0	"	" " "		2
273	0	0	"	" " "		2
295	0	0	"	" " "		2
256	0	0	"	" " "		2
275	0	1	11/15	released		
294	0	0	11/15	0;released		2
939	0	0	10/18	controls		2
901	4	4	11/15	0;released,misread		2
934	4	4	9/24	s		2
947	0		9/19	bac,stres	l	1
926	0		9/22	bact,		2
291	0		?	bacteria	l	2

Table 8c, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	spore type released	shell type
277	4		?	sporu-?	?	1
263	4		9/26		l	1
281	0	0	10/18	control		2
266	0	0	" "	" "		2
284	4		10/26	bacteria,		1
946	4	4	9/27		s	1
925	4		10/12	bacteria		1
910	3		9/25		l	1
923	4		9/17	bact,veg	sporu-?	1
938	4		10/16	bacteria		1
913	4		?	bact,4+	sporu-?	1
904	4		9/18		l	1
909	4	4	9/19	bac,stre	s	1
924	4		?	unknown	?	1
936	3		9/29	bacteria	l	1
902	4		9/28	bacteria	l	1
929	4		9/19		l	1
922	4		9/23		l,s	1
928	4		9/28	bacteria	sporu-?	1
930	4		9/23		l,s	1
908	3		9/19		l	1
906	4		10/5	bacteria		1
903	4		?	unknown	?	1
942	4		10/17	bacteria		1
905	4		9/26	bacteria		1
941	4		10/3		l	1
254	4		?	unknown	?	1
282	3		9/22	sacrifice	sporu-?	1
935	4		9/29		l	1
285	4		9/29	bacteria	sporu-?	1
259	4		10/1		l	1
290	4		?	unknown	?	1
298	3		9/19		l	1
262	4		?	unknown	?	1
296			9/26	bacteria	l,s	1
911	4		9/18	unknown	sporu-?	1
253	4		9/19		l	1
270	4		9/24	bacteria	l,s	1
920			9/17	bacteria	l	1
288	4		9/26	bacteria		1
948	2		10/7		l	1
940	4		9/29		l	1
937			9/17	bacteria	l	1
943	4		?	unknown	?	1

Table 8c, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	spore type released	shell type
933	4		9/17		l	1
914	3		9/20		l,s	1
917	3		9/19		l	1
931	2		9/19		l,s	1
untagged			10/1	bacteria	red hemo	

Table 8d. October sample, 1989 for BCD intensity before and after 60-day observation period, date and cause of mortality, sporulation and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
196	0	0	12/5	all crabs	2
194	0	0	12/5	released	2
185	0	0	12/5	" "	2
178	0	0	12/5	" "	2
163	1	1	12/5	" "	2
164	0	0	12/5	" "	2
172	0	0	12/5	" "	2
168	0	0	12/5	" "	2
167	1	1	12/5	" "	2
165	0	0	12/5	" "	2
192	0	0	12/5	" "	2
190	0	0	12/5	" "	2
175	0	0	12/5	" "	2
193	0	0	12/5	" "	2
161	0	1	12/5	" "	2
184	0		11/13	unknown	2
189	0	0	12/5	released	2
176	0	0	12/5	" "	2
195	1		12/5	sporu-?	2
158	0	0	12/5	released	2
157	0	0	12/5	" "	2
156	0	0	12/5	" "	2
155	0	0	12/5	" "	2
154	0	0	12/5	" "	2
151	0	0	12/5	" "	2
166	0	0	12/5	" "	2
152	0	0	12/5	" "	2
153	0	0	12/5	" "	2
179	0	0	12/5	" "	2
187	0	0	12/5	" "	2
182	0	0	12/5	" "	2
181	0	0	12/5	" "	2
174	0	0	12/5	" "	2
170	0	0	12/5	" "	2
160	0	0	12/5	" "	1
188	0	0	12/5	" "	1
177	4		12/5	bacteria	1
159	0	0	12/5	released	1
150	0		12/5	no final	1
191	0	0	12/5	released	1
173	0	0	12/5	" "	1

Table 8d, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
169	0	0	12/5	" "	1
171	0	0	12/5	" "	1
180	0	1	12/5	" "	1
186	0	0	12/5	" "	1
162	4		11/3	bacteria	1
183	4		11/13	unknown	2
255	0	0	12/5	released	1
256	0	0	12/5	" "	1
251	0	0	12/5	" "	1
260	0	0	12/5	" "	1
258	0	0	12/5	" "	1
262	0	0	12/5	" "	1
259	0	0	12/5	" "	1
257	0	0	12/5	" "	1
253	0	0	12/5	" "	1
254	0	0	12/5	" "	1
261	0	0	12/5	" "	1
250	0	0	12/5	" "	1
252	0	0	12/5	" "	1
263	0	15	?	no final	1
untagged			10/7	unknown	1
196	0	0	12/5	all crabs	2
194	0	0	12/5	released	2
185	0	0	12/5	" "	2
178	0	0	12/5	" "	2
163	1	1	12/5	" "	2
164	0	0	12/5	" "	2
172	0	0	12/5	" "	2
168	0	0	12/5	" "	2
167	1	1	12/5	" "	2
165	0	0	12/5	" "	2
192	0	0	12/5	" "	2
190	0	0	12/5	" "	2
175	0	0	12/5	" "	2
193	0	0	12/5	" "	2
161	0	1	12/5	" "	2
184	0		11/13	unknown	2
189	0	0	12/5	released	2
176	0	0	12/5	" "	2
195	1		12/5	sporu-?	2
158	0	0	12/5	released	2
157	0	0	12/5	" "	2
156	0	0	12/5	" "	2
155	0	0	12/5	" "	2

Table 8d, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
154	0	0	12/5	"	2
151	0	0	12/5	"	2
166	0	0	12/5	"	2
152	0	0	12/5	"	2
153	0	0	12/5	"	2
179	0	0	12/5	"	2
187	0	0	12/5	"	2
182	0	0	12/5	"	2
181	0	0	12/5	"	2
174	0	0	12/5	"	2
170	0	0	12/5	"	2
160	0	0	12/5	"	1
188	0	0	12/5	"	1
177	4		12/5	bacteria	1
159	0	0	12/5	released	1
150	0		12/5	no final	1
191	0	0	12/5	released	1
173	0	0	12/5	"	1
169	0	0	12/5	"	1
171	0	0	12/5	"	1
180	0	1	12/5	"	1
186	0	0	12/5	"	1
162	4		11/3	bacteria	1
183	4		11/13	unknown	2
255	0	0	12/5	released	1
256	0	0	12/5	"	1
251	0	0	12/5	"	1
260	0	0	12/5	"	1
258	0	0	12/5	"	1
262	0	0	12/5	"	1
259	0	0	12/5	"	1
257	0	0	12/5	"	1
253	0	0	12/5	"	1
254	0	0	12/5	"	1
261	0	0	12/5	"	1
250	0	0	12/5	"	1
252	0	0	12/5	"	1
263	0	15	?	no final	1
untagged			10/7	unknown	1

Table 8e. November sample, 1989 for BCD intensity before and after 60-day observation period, date and cause of mortality, sporulation and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

	intensity of infection				
Crab #	initial	final	date of death	cause of death	shell type
1289	0	0	1/8	all crabs	2
1285	0	0	1/8	released	2
1864	0	0	1/8	upon	2
1283	0	0	1/8	final	2
1854	0	0	1/8	screen	2
1894	0	0	1/8		2
1297	0	0	1/8		2
1282	0	0	1/8		2
1271	0	0	1/8		2
1299	1	1	1/8		2
1294	0	0	1/8		2
1869	0	0	1/8		2
1862	0	0	1/8		2
1266	0	0	1/8		2
1281	0	0	1/8		2
1863	0	0	1/8		2
1870	0	0	1/8		2
1865	0	0	1/8		2
1298	1	1	1/8		2
1288	0	0	1/8		2
1278	0	0	1/8		2
1277	0	0	1/8		2
1878	0	0	1/8		2
1859	0		?	no final	2
1295	0	0	1/8		2
1886	0	0	1/8		2
1850	0	0	1/8		1
1879	0	0	1/8		2
1267	0	0	1/8		2
1287	0	0	1/8		2
1268	0	0	1/8		2
1293	0	0	1/8		2
1198	2		?	no final	2
1269	0	0	1/8		2
1264	0	0	1/8		2
1270	0	0	1/8		2
1853	0	0	1/8		2
1876	0	0	1/8		2
1874	0	0	1/8		2
1871	0	0	1/8		2
1291	0	0	1/8		2

Table 8e, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
1276	0	0	1/8		2
1296	0	0	1/8		2
1888	0	0	1/8		2
1872	0		?	no final	2
1265	0	1	1/8		2
1292	0	0	1/8		2
1275	1	1	1/8		2
1881	0	0	1/8		2
1274	0	0	1/8		2
1896	0	0	1/8		2
1873	1	1	1/8		2
1898	0	0	1/8		2
1885	0	0	1/8		2
1875	0	0	1/8		2
1897	0	0	1/8		2
1893	0	0	1/8		2
1279	0	0	1/8		1
1290	3	4	1/8		1
1280	0	0	1/8		1
1882	0	0	1/8		1
1272	0	0	1/8		1
1273	0	0	1/8		1
1887	0	0	1/8		1
1286	0	0	1/8		1
untagged	0	18	1/8		1
males	0	18		no final	1
" "	0	18		screen	1
" "	0	18		" "	1
" "	0	18		" "	1
" "	0	18		" "	1
" "	0	18		" "	1
" "	0	18		" "	1
" "	0	18		" "	2
" "	0	18		" "	2
" "	0	18		" "	2
" "	0	18		" "	2
" "	0	18		" "	2
" "	0	18		" "	2
" "	0	18		" "	2
" "	0	18		" "	2

Table 8f. December sample, 1989 for BCD intensity before and after 60-day observation period, date and cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
876	0	0	54	only 28	1
884	0	0	54	crabs	1
864	0	0	54	taken this	3
898	0	0	54	month--	2
871	0	0	54	fishing	2
890	0	0	54	condition	3
874	0	0	54	bad and	2
866	0	0	54	pots not	1
878	0	0	54	working	2
852	0	0	54	effective	2
855	0	0	54	crabs also	2
868	0	0	54	seen to	2
896	0	0	54	be more	2
882	0	0	54	spread	2
889	0	0	54	out or	1
857	0	0	54	less	2
867	0	0	54	active	2
893	0	0	54		1
854	0	0	54		2
856	0	0	54		2
881	0	0	54		3
869	0	0	54		2
861	0	0	54		2
853	0	0	54		1
873	0	0	54		2
892	0	0	54		3
888	0	0	54		2
863	0	0	54		2

Table 8g. February sample, 1990 for BCD intensity before and after 60-day observation period, date adn cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
no tag	0		2/5	no final	
no tag	0		2/6	screen	
345	0		4/2	coldshock	2
326	0	0	4/2	all crabs	2
342	0	0	3/30	surviving	2
315	0	0	3/30	until	2
312	0	0	3/30	final	2
313	0	0	3/30	screen	2
335	0	0	4/2	released	2
325	0	0	3/30		2
331	0	0	3/30		2
320	0	0	3/30		1
300	1	3	4/2		2
346	0	0	3/30		1
329	0	0	3/30		1
311	1	1	3/30		1
336	0	1	4/2		2
306	0		4/2	molted	2
334	0	0	4/2		2
332	0	0	4/2		2
339	1	3	3/30		1
330	0	0	4/2		2
347	0	0	3/30		1
324	0	0	4/2		2
344	0	0	4/2		2
316	0	0	4/2		2
308	0	0	4/2		2
341	0	0	3/30		1
343	0	0	4/2		2
220	0	0	4/2		1
224	0	0	4/2		2
201	0	0	4/2		2
215	0	0	3/30		1
245	0			no final	2
231	0	0	3/30		1
200	0	0	4/2		2
238	0	0	4/2		2
249	0	0	4/2		2
213	0	0	3/30		2
233	0	0	4/2		
221	0	0	4/2		2

Table 8g, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
232	0	0	4/2		2
209	0	0	4/2		1
246	0	0	4/2		2
210	0	0	4/2		2
237	0	0	4/2		2
208	0	0	4/2		2
236	0	0	3/30		1
212	0		3/19	molted	2
242	0			no final	1
286	0			no final	
202	0	0	3/30		1
205	0	0	3/30		2
222	0	0	3/30		1
218	0	0	3/30		2
214	0	0	3/30		1
207	1	1	3/30		1
219	0	0	3/30		1
203	0	0	4/2		2
239	0	0	3/30		1
217	4		3/19	lethargy	1
235	1		3/26	unknown	1
228	0	2	4/2		2
no tag	1			no final	
mortality	0			no final	1
248	0	0	3/30		1
230	0	0	3/30		2
244	0	0	3/30		1
204	4		3/7	sporu?	1
241	0	0	3/30		1
225	0	0	3/30		1

Table 8h. March sample, 1990 for BCD intensity before and after 60-day observation period, date and cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
1	1	3/2			2
2	4	3/2			2
3	0	3/2			2
4	2	3/2			2
5	1	3/2			2
6	4	3/2			2
7	0	3/2			1
8	1	3/2			1
9	1	3/2			1
10	1	3/2			1
11	1	3/2			2
12	1	3/2			2
13	0	3/2			?
14	1	3/2			1
15	1	3/2			1
16	0	3/2			1
17	0	3/2			1
18	0	3/2			1
19	1	3/2			2
20	0	3/2			1
21	1	3/2			2
22	0	3/2			2
23	0	3/2			1
24	0	3/2			2
25	0	3/2			2
26	0	3/2			2
27	0	3/2			2
28	1	3/2			2
29	1	3/2			2
30	0	3/2			2
31	0	3/2			2
32	1	3/2			2
33	0	3/2			1
34	1	3/2			1
35	0	3/2			1
36	1	3/2			1
37	0	3/2			1
38	0	3/2			1
504	0	0	5/1		1
502	0		4/23		2
589	0	0	5/1		1

Table 8h, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
548	0	0	5/1	all HOH	2
582	0	0	5/1	failures	2
526	0	0	5/1	= 5/8	2
506	0	0	5/1		2
527	1	3	5/1	OHO fail	2
531	0	0	5/1	bact 5/17	1
585	0	0	5/1		1
546	0	0	5/1		1
590	4	4	5/1		1
535	0	0	5/1		1
519	0	0	5/1		2
537	0	0	5/1	OHO fail	2
561	0	0	4/23		2
510	0	0	5/1		1
541	0	0	5/1	OHO fail	2
557	0	0	5/1		1
584	0	0	5/1		1
581	0	0	5/1		1
564	0	0	4/23		2
550	0	0	5/1	OHO fail	2
529	0	0	5/1	bact 6/6	1
528	0	0	5/1	OHO fail	1
583	0	0	5/1		1
559	0	0	5/1		1
562	0	0	5/1	OHO fail	1
594	0	3	5/1		1
518	0	0	5/1	OHO fail	1
574	0	0	5/1	bact 5/11	2
555	0	0	4/23	OHO fail	2
556	0	0	4/23	OHO fail	2
566	0	1	4/23	OHO fail	2
505	0	0	5/1		1
512	0	0	5/1	OHO fail	1
515	0		4/23		2
503	0	0	5/1		1
597	0	4	4/23		2
513	0	0	5/1		1
544	0	0	5/1		1
595	0	0	5/1		1
573	0	0	5/1		2
588	0	0	5/1		1
569	0		5/1	spor?5/17	1
516	0	0	5/1	OHO fail	2
587	0		?		1

Table 8h, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
571	0	0	5/1	bact?5/17	1
514	0	0	5/1		1
500	0	0	5/1		2
509	1	6	5/1	OHO fail	1
575	0	0	5/1		2
596	0	0	5/1		2
599	0	0	5/1		2
578	0	0	5/1		1
572	0	1	5/1		2
530	0	0	5/1		1
576	0	1	5/1	spor?5/18	2
539	0	0	5/1		1
525	0	0	5/1	bact 6/6	1
567	0	0	5/1		2
533	0	0	5/1		1
579	0	0	5/1		2
551	0	0	5/1		1
507	0	0	5/1		2
536	0	0	5/1		2
532	0	0	5/1		1
524	0	0	5/1		1
540	1	0	5/1		1
534	0	0	5/1		1
568	0	0	5/1		1
517	0	0	5/1	bact 5/29	2
586	0	0	5/1	bact 5/29	1
522	0	0	5/1		1
545	0	0	5/1		1

Table 8i. April sample, 1990 for BCD intensity before and after 60-day observation period, date and cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

	intensity of infection		date of	cause of	shell
Crab #	initial	final	death	death	type
1920	0	0	6/7		1
1978	0	0	6/7		1
1966	0	0	6/7		1
1942	0	0	6/7		1
1959	0	0	6/7		1
1905	0	0	6/7		1
1914	0	1	6/7		1
1968	0	0	6/7		1
1900	0	0	6/22	Ciliates	1
1963	0	0	6/7		1
1947	0	0	6/7		1
1981	0		6/7		1
1965	0	0	6/7		1
1903	0	0	6/7		1
1975	0	0	6/7		1
1950	0	1	6/7		1
1960	0	0	6/7		1
1901	0	0	6/7		1
1925	0	0	6/7		1
1902	0	4	6/7		1
1907	0	0	6/7		1
1977	0	0	6/7		1
1976	0	0	6/7		1
1916	0	0	6/7		1
1933	0	0	6/7		1
1908	0	0	6/7		1
1932	0	0	6/7		1
1935	0	0	6/7		1
1904	0	0	6/7		1
1988	0			unknown	1
1944	0	3	6/7		1
1961	0	0	6/7		1
1926	0	0	6/7		1
1929	0	0	6/7		1
1946	0		5/20	Bacteria	1
1936	0	0	6/7		1
1927	0		5/19	unknown	1
1931	0		5/29	Bact, cil	1
1930	0	0	6/7		1
1934	0	0	6/7		1
1964	0		5/29	bact,cili	1

Table 8i, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
1923	0	4	6/7	bacteria	1
1945	0	0	6/7		1
1955	0	0	6/7		1
1956	0	0	6/7		1
1952	0		6/7		1
1971	0	0	6/7		1
1908	0		6/7		1
1954	0	0	6/7		1
1912	0	0	6/7		1
1906	0	0	6/7		1
1919	0		6/7		1
1921	0	0	6/7		1
1915	0	0	6/7		1
1922	0	0	6/7		1
1949	0	0	6/7		1
1974	0	0	6/7		1
1957	0	0	6/7		1
1939	0	0	6/7		1
1970	0	0	6/7		1
1913	0	0	6/7		1
1917	0	0	6/7		1
1938	0	0	6/7		1
1948	0	0	6/7		1
1958	0	0	6/7		1
1924	0	0	6/7		1
1940	0	0	6/7		1
1993	0	0	6/7		1
1972	0	0	6/7		1
1928	0	4	6/7		1
1967	0	0	6/5	Ciliates,	1
451	0	0	6/7		
1951	0	0	6/7		1
1962	0	0	6/7		
1918	0	0	6/7		
1909	0	0	6/7		1
1985	0	0	6/7		1
1911	0		4/23	unknown	1
1937	0		5/19	Bacteria	1

Table 8j. May sample, 1990 for BCD intensity before and after 60-day observation period, date and cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
111	0	0	6/28		2
489	0	0	6/28	released	2
145	0	1	6/28	released	2
143	0	0	6/28		1
144	0	0	6/28		2
100	0	9	6/6	bacteria	1
133	0	0	6/28	released	1
137	0	0	6/28	h20	2
126	0		6/12	failure	2
123	0		6/12	" "	2
140	0	0	6/28		2
121	0	0	6/28		2
105	0	0	6/28		2
118	0		6/5	H20 failr	2
119	0	0	6/28		2
141	0	0	6/28		2
452	0	0	6/28	released	2
451	0	0	6/28	released	2
471	0	0	6/28	released	2
479	0		6/5	H20 failr	1
110	0	0	6/28	released	2
107	0	0	6/28		2
145	4		?	unknown	1
131	1	4	6/28	released	1
147	0	0	6/28	released	1
104	0	0	6/28		2
142	0	0	6/28	released	2
114	0		6/5	H20 failr	1
103	0		6/12	H20 failr	2
175	0		?	unknown	2
148	0	0	6/28	released	2
486	0	0	6/12	H20 failr	2
480	0	0	6/28	released	1
490	0	0	6/5	H20 failr	1
116	0		6/5	H20 failr	2
113	0	0	6/28		2
112	0	0	6/28	released	1
106	0		?	unknown	1
130	2		6/12	H20 failr	2
102	0	0	6/28	released	1
117	0	0	6/28		2

Table 8j, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
109	0		5/24	bact(5/24	2
127	3		5/30	Sporu-?	2
115	1	3	6/28	released	1
493	0	0	6/28	released	2
454	0		6/5	H20 failr	2
496	0	0	6/28	released	2
450	0		6/28	unknown	2
461	0		6/5	H20 failr	2
463	0		?	unknown	2
488	0	0	6/28	released	2
459	0		6/5	H20 failr	2
469	0	0	6/28	released	2
485	0		6/5	H20 failr	2
481	0		7/17	bact,micr	2
487	0	0	6/28	released	2
478	0	1	6/28	released	2
495	0		6/5	H20 failr	1
472	0	0	6/28	released	2
457	0		6/5	H20 failr	2
460	0	0	6/28	s?	2
456	0	0	6/28	released	1
453	0	0	6/28	released	2
458	0	0	6/28	released	2
482	0	0	6/28	bad slide	2
484	1	3	6/28	released	1
464	0	0	6/28	released	2
467	0		6/5	H20 failr	2
465	0		6/5	H20 failr	2
492	0	0	6/28	released	2
491	0	0	6/28	released	2
455	0		5/30	bact(5/30	2
477	0		?	unknown	2
475	3	4	6/28	?prespore	1
474	0	0	6/28		2
466	1	0	6/28"-"	molted	2
483	4		6/5	H20 failr	1
476	0		6/5	H20 failr	1
462	0		6/5	H20 failr	2
470	0		6/5	H20 failr	2
499	0	0	6/28	"Bob"	2
no tag	0	4	6/28	no initial	
124	0	0	6/28	released	2
146	0		6/5	H20 failr	2
120	0	0	6/28	released	2

Table 8j, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
122	0		6/5	H20 failr	2
134	4		?	unknown	2
135	0		6/5	H20 failr	2
136	0		6/5	H20 failr	2
132	0	0	6/28	released	2
101	0		6/5	H20 failr	2
138	0		6/5	H20 failr	3
128	0		6/5	H20 failr	2
129	0	0	6/28	released	2
1	2	2	5/10	shock	1
2	0	0	5/10	shock	1
708	0	0	6/5	H20 failr	1
722	1	3	6/28	released	1
716	0	0	6/28	released	1
715	0		6/5	H20 failr	1
473			6/5	H20 failr	1
498			6/5	H20 failr	1
497				no record	2

Table 8k. June sample, 1990 for BCD intensity before and after 60-day observation period, date and cause of mortality for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death
652	1	4	7/24	sporu- sm
651	0	4	8/14	sporu-sm
672	0	0	8/16	0;physiol
655	2	4	8/7	4+,releas
661	0		8/2	sporu-? d
678	4		7/6	unknown
659	0	0	8/16	0;physiol
687	0	3	8/7	3+,releas
656	0		8/16	1+ relea
680	1		7/29	bact,veg
653	4	4	7/12	small
684 ?0	0	0	8/16	0;physiol
671	0	0	8/16	0,release
698	0	1	8/7	1+ or sma
693	0	0	8/16	0, releas
686	0	1	8/7	1+ or sma
682	0		8/2	4+(7/16)
696	0	0	8/16	0;physiol
676	0	4	8/16	4+,releas
683	0	0	8/16	0,release
685	3		?	unknown
670	0		6/25	unknown
663	0	0	8/16	0;physiol
666	0	0	8/16	0;physiol
689	0	0	8/16	0;physiol
690				where is
660	0	5	7/24	small
694	0	0	8/16	0;physiol
677	0	0	8/16	0;physiol
675	0	4	8/7	4+,prespo
679	0	0	8/16	0;physiol
681	0		8/7-,8/16	"-" relea
665	0	0	8/16	0;physiol
691	1	1	8/7	1+, relea
697	3	4	7/19	small
688	0	0	8/16	0;physiol
662	0		8/7-,8/16	released
654	4		7/16	Sporu-?
664	1		7/22	sporu-?
673			8/16	bad slide
669	0		8/7-,8/16	"-" relea

Table 8k, continued.

Crab #	intensity of infection initial	final	date of death	cause of death
667	0	0	8/16	0;physiol
692	0	0	8/16	0;physiol
668	0	2	8/7	2+,releas
658	1		8/16	BACT
699	3	4	7/22	small
657	0	0	8/7-,8/16	"-" relea
674	0	0	8/7-,8/16	released
695	1	4	8/7	4+
650	3	4	8/7	4+ final,
99	0	0	8/16	0;physiol
91	0		8/7-,8/16	released
71	0		8/7-,8/16	released
74	0		8/7-,8/16	released
60	0	3	8/7	2-3+,smal
68	0		8/7-,8/16	released
73	0	4	8/16	8/7"4+,8/
64	0		8/7-,8/16	released
86	0	0	8/16	0;physiol
80	0	0	8/16	0;physiol
89	0		6/22	unknown
79	0		6/25	unknown
55	0	0	8/16	0;physiol
77	0		8/7-,8/16	released
72	0		8/7-,8/16	released
54	0		8/7-,8/16	released
56	0		7/14	sporu-?
94	0		8/7-,8/16	released
88	0		8/7-,8/16	released
59	0		8/7-,8/16	released
66	4		7/17	veg,large
78	0		8/7-,8/16	released
96	0	4	8/2	small
51	1	4	7/29	small
90	0		8/7-,8/16	released
97	0		8/7-,8/16	released
75	0		8/7-,8/16	released
81	3	4	8/16	released
69	0	0	8/16	0;physiol
53	0		7/16	sporu-?
58	0		6/25	unknown
85	0		6/25	unknown
52	0	0	8/16	0;physiol
76	0	0	8/16	0;physiol
84	0		8/7-,8/16	released

Table 81. July sample, 1990 for BCD intensity before and after 60-day observation period, date and cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
514	0	0	7/30	released	2
519	0	0	7/30	" "	2
541	0	0	7/30	" "	2
512	0	0	7/30	" "	2
538	0	0	7/30	" "	2
518	0	0	7/30	" "	2
524	0	0	7/30	" "	2
521	0	0	7/30	" "	2
528	0	0	7/30	" "	2
506	0	0	7/30	" "	2
533	4	4	7/16	4+ mort	2
543	0	0	7/30	released	2
510	0	0	7/30	" "	2
509	0		7/13	unknown	2
535	0	0	7/30	released	2
526	0	0	7/30	" "	2
517	0	0	7/30	" "	2
520	0	0	7/30	" "	2
531	0	0	7/30	" "	2
511	0	0	7/30	" "	2
508	0	0	7/30	" "	2
507	0	0	9/20	" "	1
530	0	0	9/20	" "	2
532	0	0	7/30	" "	2
522	0	0	7/30	" "	2
505	0	0	7/30	" "	2
545	0	0	7/30	" "	2
523	0	0	7/30	" "	2
516	0	0	7/30	" "	2
513	0	0	7/30	" "	2
502	0	0	7/30	" "	2
548	0	0	7/30	" "	2
544	0	0	7/30	" "	2
547	0	0	7/30	" "	2
529	0	0	7/30	" "	2
534	0	0	7/30	" "	2
546	4	4	7/30	4+ releas	2
525	0	0	7/30	released	2
500	0	0	7/30	" "	2
536	0	0	7/30	" "	2
537	0	0	7/30	" "	2

Table 81, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
501	0	0	7/30	" "	2
800	0	0	7/30	" "	2
504	4	5	7/22	small	1
503	4	5	7/22	small	1
549	4		8/13	sporu-?	1
206	4	6	7/21	large,bac	1
319	4	4	7/16	4+	1
245	4	4	9/20	4+	1
216	2		9/12	bact,veg,	1
323		5	8/13	no initia	1
335	4	4	7/30	4+	1
527	1		8/3	sporu-?,v	1
234	0	0	9/20	released	2
243	0	0	7/16	released	2
139	0	0	9/20	released	2
539	4	4	9/20	4+	1
542	4	4	7/16	blackmat,	2
342	0	0	9/20	released	2
247	4	4	9/20	released	1
223	0	0	9/20	released	2
226	4	7	7/17	large/sma	1
325	3	5	7/18	small	2
229	4	6	8/24	large	1
816	0	0	7/30	released	2
814	0	0	7/30	" "	2
841	0	0	7/30	" "	2
817	0	0	7/30	" "	2
828	0	0	7/30	" "	2
806	0	0	7/30	" "	2
847	0		?	unknown l	2
813	0	0	9/20	released	2
801	0	0	7/30	" "	2
823	0	0	7/30	" "	2
821	0	0	7/30	" "	2
807	0	0	9/20	" "	1
830	0	0	7/30	" "	2
812	0	0	7/30	" "	2
839	0	0	7/30	" "	2
842	0	0	7/30	" "	2
802	0	0	7/30	" "	2
805	0	0	7/30	" "	2
832	0	0	7/30	" "	2
831	0	0	7/30	" "	2
845	0	0	7/30	" "	2

Table 81, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
815	0	0	7/30	"	2
808	0	0	7/30	"	2
810	0	0	7/30	"	2
820	0	0	7/30	"	2
840	0	0	7/30	"	2
819	0	0	7/30	"	2
829	0	0	7/30	"	2
803	0	0	7/30	"	2
835	0	0	7/30	"	2
848	0	0	7/30	"	2
843	0	0	7/30	"	2
838	0	0	7/30	"	2
822	0	0	7/30	"	2
826	0	0	7/30	"	2
804	0	0	7/30	"	2
846	0	0	7/30	"	2
844	0	0	7/30	"	2
825	0	0	7/30	"	2
849	0	9	7/30	bacteria	2
809	0	0	7/30	released	2
833	0	0	9/20	"	2
827	1	0	7/30	"	1
837	0	0	9/20	released	2
834	4	4	7/16	4+	1

Table 8m. August sample, 1990 for BCD intensity before and after 60-day observation period, date and cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
1063	0	0	10/24	released	2
1069	0	0	10/24	" "	2
1066	0	0	10/24	" "	2
1581	0	0	10/24	" "	2
1052	0	0	10/24	" "	2
1062	0	0	10/24	" "	2
1073	2		10/7	water fai	1
1084	0	4	10/24	released	1
1075	0		10/7	water fai	1
1088	4		10/7	"	1
719	0		10/7	"	1
718	0		10/7	"	1
714	1		10/7	"	1
725	3	4	8/13	small	1
748	0		10/7	water fai	1
736	0	0	10/24	released	1
700	0	0	10/24	" "	1
745	0		10/7	water fai	1
720	0		10/7	"	1
733	4		10/7	"	1
738	1		10/7	"	1
746	4	4	8/6	small	2
709	1		10/7	water fai	2
730	4	4	8/21	small	1
705	0		10/7	water fai	1
703	2		10/7	"	1
743	4	4	8/21	small/bac	1
742	1		10/7	water fai	2
735	0	0	10/24	released	1
762	0	0	10/24	" "	1
790	0	0	10/24	" "	1
772	4	0	10/24	" "	1
711	3	4	8/8	small	1
726	1	0	10/24	released	1
751	4	4	8/7	small/lat	1
759	0	0	10/24	released	2
729	0		10/2	bacteria	2
732	4	4	10/24	released	1
758	4	4	10/24	released	1
794	0	0	10/24	released	1
799	0		8/4	bact,micr	1

Table 8m, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
723	2		9/5	unknown	2
791	2	4	10/24	released	1
771	4	4	8/16	small	1
765	3		8/15	?sporu-,	1
753	4	4	8/13	small	1
713	0		8/16	"-"	1
793	0	0	10/24	released	1
752	0	0	10/24	" "	2
710	0	0	10/24	" "	1
747	1	1	10/24	" "	1
789	4		?	Unknown	1
702	0	0	10/24	released	2
787	3		8/21	sporu-?,b	1
786	0	0	10/24	released	2
777	4		9/5	unkown	1
797	4	4	8/13	small	1
794		0	10/24	first scr	1
749	4		10/7	water fai	1
795	4		8/2	large	1
781	4		?	unknown	1
728	4		8/21	sporu-?,b	1
741	0		8/16	bact	2
756	1	4	8/6	small	1
769			10/7	water fai	1
780	1	4	8/6	small/lat	1
785	1		10/7	water fai	1
739	0	0	10/24	released	2
706	0	0	10/24	" "	1
779	0	1	10/24	" "	1
717	4	4	8/16	small/lat	1
774	3	0	10/24	released	1
750	4		8/13	unknown/b	2
764	0	0	10/24	released	2
767	1	0	10/24	" "	2
773	4	4	8/4	small	2
776	1	1	10/24	released	1
737	0	0	10/24	released	1
731	4	4	8/3	small	1
783	3		9/25	veg,paran	1
740	4		?	unknown	1
763	0	0	10/24	released	2
775	0	0	10/24	" "	2
757	3		9/12	veg,bact,	1
760	4	4	8/2	small	

Table 8m, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
796	4		8/4	sporu-?,veg,bac,mi	
792	4		9/12	veg,bact,sporu?	
782	0	0	10/24	released	
713	0	1	10/24	released	
712	4		?	unknown	

Table 8n. September sample, 1990 for BCD intensity before and after 60-day observation period, date and cause of mortality, and molt stage (1 for new, 2 for old) for Tanner crabs from Auke Bay, Alaska.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
191	4	4	10/24	released	1
185	0	0	10/24	" "	1
182	0		?	unknown	1
190	1	3	10/24	released	1
156	3	3	10/24	" "	1
177	0	0	10/24	" "	1
167	0	0	10/24	" "	2
195		0	10/24	?initial	2
198	0	0	10/24	released	2
197	0	0	10/24	released	2
187		0	10/24	?initial	2
154	0	0	10/24	released	2
178	0	0	10/24	" "	2
165	0	0	10/24	" "	2
155	0	0	10/24	" "	2
175	4		?	unknown	1
186	0	0	10/24	released	2
183	0	0	10/24	" "	2
188	0	0	10/24	" "	2
169	0	0	10/24	" "	2
194	3		10/4	bact/unkn	1
161	0		?	unknown	2
172	0	0	10/24	released	2
171	4	5	10/6	small	1
150		0	10/24	no initia	2
152	0	0	10/24	released	2
166	0	0	10/24	" "	2
160	1	1	10/24	released	2
157	0	0	10/24	" "	2
170	0	0	10/24	" "	2
192	0	0	10/24	" "	2
168	0	0	10/24	" "	2
159	0	0	10/24	" "	2
164	0	0	10/24	" "	1
193	4		9/24	large	1
158	0		10/21	?sporu?,	2
184	1		?	unknown	1
173	0	0	10/24	released	2
163	0	0	10/24	" "	2
180	1	2	10/24	released	1
199	0	0	10/24	released	2

Table 8n, continued.

Crab #	intensity of infection initial	final	date of death	cause of death	shell type
189	4		10/6	unknown	1
174	1		10/24	poor slid	2
153	2	4	10/24	released	1
176	1	3	10/24	" "	2
181	0		10/24	unknown	2
151	4		10/21	?sporu-,	1
196	0	0	10/24	released	2
106	4		10/21	?sporu-,	2
114	0	0	10/24	released	2
100	0	0	10/24	" "	2
107	4	4	10/24	" "	1
105	0	0	10/24	" "	1
144	4		9/25	large	1
137	4		9/24	large	1
141	0	0	10/24	released	1
134	0	0	10/24	" "	2
145	0	0	10/24	" "	2
148	0	0	10/24	" "	2
124		0		no initial	
129		0		screens	
132		0		" "	
133		0		" "	
139		0		" "	
142		0		" "	